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THE COMPARATIVE EARLY EMBRYOLOGY OF THE OLIGOCHAETA, HIRUDINEA AND ONYCHOPHORA

D. T. ANDERSON

School of Biological Sciences, University of Sydney

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Synopsis

The primitive, large, yolky eggs of tubificid and lumbricid oligochaetes and glossiphoniid leeches undergo a modified spiral cleavage yielding a yolky blastula. The pattern of presumptive areas in the blastula wall is interpretable as a modification of the polychaete presumptive area pattern, in association with increased yolk. The same pattern of presumptive areas is retained in the secondarily microlecithal embryos of lumbricid oligochaetes, gnathobdellid and pharyngobdellid leeches and probably other clitellates, in spite of further cleavage modifications and of embryonic adaptations to albumenotrophy.

The primitive, large, yolky eggs of ovoviviparous Onychophora undergo centrolecithal cleavage, setting out a blastoderm around a central yolk mass. In viviparous non-placental species, although yolk is absent and cleavage is modified, a similar blastoderm is usually formed around a central cavity. In viviparous placental species, cleavage is highly specialized, yielding a small blastodermic vesicle at the end of a placental stalk and plate.

The blastodermal presumptive areas of Onychophora, like those of clitellates, retain a constant pattern in spite of secondary yolk loss and its attendant modifications. Taking into account the events of gastrulation, the blastodermal presumptive area pattern of Onychophora is interpretable as a modification of the primitive clitellate presumptive area pattern, in association with a further increase in yolk.

It is suggested that a clitellate-like mode of early embryonic development must have characterized the ancestors of the Onychophora, and that these ancestors were members of the spiral cleavage assemblage and were probably paraphyletic with the ancestors of annelids.

INTRODUCTION

Following a review of the embryology of polychaete annelids by Anderson (1966*a*), the embryonic development of clitellate annelids and the modifications which they show as compared with polychaetes now become a subject of renewed interest. While the development of yolky polychaete eggs always retains traces of a primitive mode of development from a small, microlecithal egg through a planktotrophic trochophore and metamorphosis, that of clitellates shows no such traces. As is well known, both oligochaetes and hirudineans deposit their eggs in a cocoon in which development proceeds directly to hatching as a miniature adult with numerous segments. Primitively, the embryo develops from a relatively large yolky egg, the cleavage, gastrulation and organogeny of which are adapted to the presence of voluminous yolk reserves. Secondly, in the majority of clitellate families, the egg is small and microlecithal and the embryo is adapted to feeding on the albuminous contents of the cocoon. The nature of both types of modification has hitherto yielded no clue as to the possible marine antecedents of primitive clitellate development or the derivation of the secondary yolkless from the primitive yolky condition. When the data of clitellate embryology are reexamined in the light of new understanding of polychaete development, however, solutions to both of these problems can be proposed, as will be shown below.

Primitive clitellate development exemplifies a particular modification of spiral cleavage development to voluminous yolk and the production of a metamerically segmented annelid. The embryonic development of Onychophora is also primitively adapted to voluminous yolk, and to the formation of a metamerically segmented animal with a number of annelid-like features. In the absence of "spiral cleavage" features in onychophoran development, the

derivation of Onychophora from a spiral cleavage ancestry has hitherto proved elusive of recognition (Manton, 1965) but, as will also be shown below, pursuance of the comparison in an appropriate way reveals the development of the Onychophora, not only as a modification of spiral cleavage development, but also as one with unmistakable clitellate-like antecedents.

It is true that certain difficulties attend this line of argument. The descriptive data on the embryology of the Oligochaeta, Hirudinea and Onychophora are incomplete in many critical ways and few of the available papers embody results obtained by modern histological techniques. Among the oligochaetes for example, *Tubifex rivulorum* and *Rhynchelmis limosella* are the only species with large, yolky eggs whose development has been adequately described (Schmidt, 1922; Penners, 1922, 1924; Swetloff, 1923b; Meyer, 1929), all other investigations being concerned with the specialized embryos of naidids, *Bdellodrilus* and earthworms. The same situation obtains for the Hirudinea, where again only two forms with large yolky eggs, *Glossiphonia* and *Theromyzon*, have been investigated (Schliep, 1914; Schmidt, 1917, 1925a), the remaining descriptions being confined to specialized piscicolid, gnathobdellid and pharyngobdellid development. The Onychophora present a further difficulty in that nothing is known of the development of the large, yolky eggs of oviparous onychophorans and only a little of the yolky eggs of ovoviviparous species (Sheldon, 1888, 1889a; Evans, 1902). On the other hand, the specialized development of the viviparous Onychophora, especially as described for *Peripatopsis* by Manton (1949), provides many inferences which are of value for comparative purposes, and permits a picture to be drawn of clitellate and onychophoran development which holds much of interest for the phylogeneticist and epigeneticist.

CLEAVAGE

Primitive cleavage in the Oligochaeta

In *Tubifex* and *Rhynchelmis*, the spiral pattern of cleavage is retained, but is modified in ways related to the magnitude of the large, yolky egg and its development in a protective cocoon. The nature of these modifications has hitherto been obscured by employment of the Wilsonian system of enumeration of spiral cleavage (Wilson, 1892), which places emphasis on the integrity of quartettes and individuality of single blastomeres and their fate. If, as for polychaetes (Anderson, 1966a), attention is alternatively directed towards the formation and subsequent fate of presumptive areas in the wall of the blastula, the oligochaete response to yolk is much more clearly discerned.

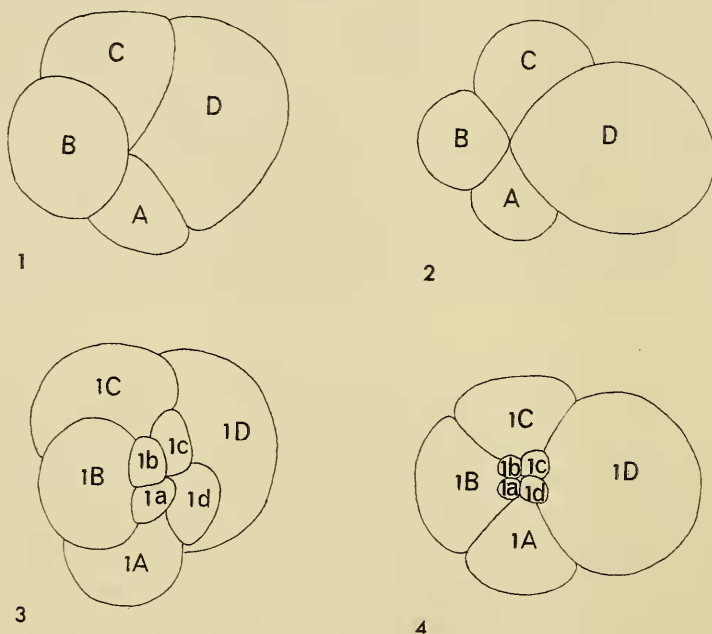
Both in *Tubifex* (Penners, 1922, 1924) and *Rhynchelmis* (Vedjovsky, 1886, 1888-92; Bergh, 1890; Penners, 1922; Swetloff, 1923b), the first two cleavage divisions are at right angles along the animal-vegetal axis of the egg, the next four perpendicular to this axis and, except for a precocious onset of bilateral cleavage in the D-quadrant, successively dextrotropic, laeotropic, dextrotropic and laeotropic in the classical spiral cleavage sequence. In association with the large size of the egg, 300-500 μ in diameter in *Tubifex*, 1000 μ in *Rhynchelmis*, the first division is very unequal, AB being much smaller than CD, and the second division maintains this inequality in the D-quadrant, D remaining large, while C is only a little larger than A and B (Figs 1, 2).

In polychaetes with yolky eggs, where the D cell is also disproportionately large, much of it is subsequently segregated into a ring of large ectoteloblasts and a pair of large M-cells within the ring, forming a growth zone which gives rise to several trunk segments before the yolk reserves of the embryo are exhausted (Anderson, 1966a). The large D cell in primitive oligochaetes is a developmental adaptation subserving a similar function. In *Tubifex*, for example, 34-38 segments are formed before the embryo hatches from the cocoon.

The orientation of the four quadrants in *Tubifex* at the 4-cell stage is as in polychaetes, D being dorsal, and A, B and C left-lateral, ventral and right-lateral respectively relative to the anteroposterior axis of the embryo, though there

is a change in quadrant orientation as development proceeds further. In *Rhynchelmis* at the 4-cell stage, on the other hand, the large D cell is posterior in position, with A on the left, B anterior and C on the right. Cleavage in *Tubifex* finally results in displacement of B-quadrant cells to an anterior and D-quadrant cells to a posterior position as described below. In *Rhynchelmis* quadrant reorientation, presumably through a redistribution of ooplasm, manifests itself in the egg.

Associated with the large size of the D cell, early cleavage after the 4-cell stage in *Tubifex* and *Rhynchelmis* differs from that of polychaetes in a marked tendency to precocious division of the D-quadrant cells, and to a lesser extent of the C-quadrant cells, so that the cells of each quartette cut off from the stem cells at the 3rd and subsequent divisions are formed in succession, the D-quadrant leading, the A and B quadrants lagging.

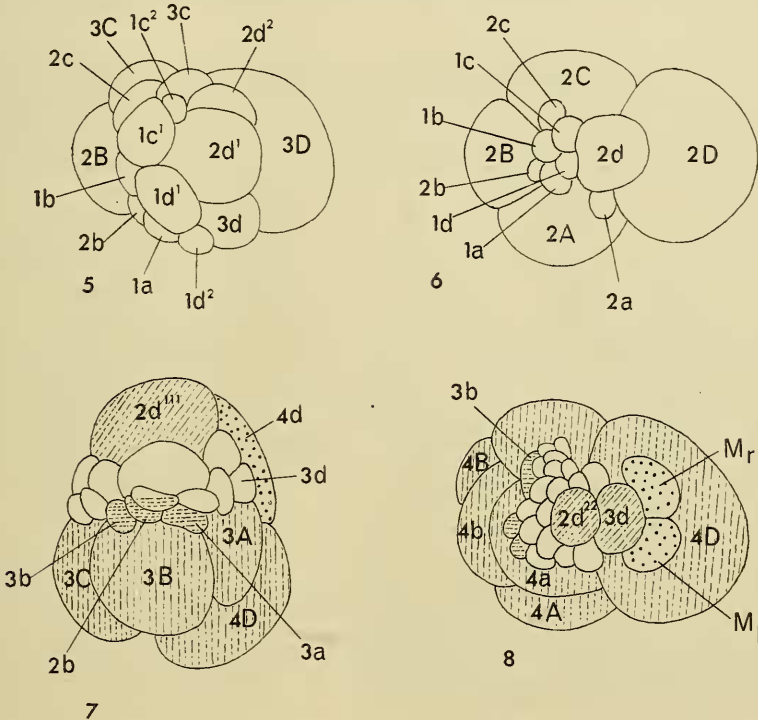


Figs 1-4.—1, *Tubifex*, 4-cell stage, anterior view with dorsal surface on the right. 2, *Rhynchelmis*, 4-cell stage, dorsal view with posterior end on the right. 3, *Tubifex*, 8-cell stage, anterior view, 4, *Rhynchelmis*, 8-cell stage, dorsal view. (After Penners, 1922; Swetloff, 1923b.) The key to the shading conventions employed in Figs 1-33 is provided by Fig. 13.

At the 3rd cleavage division (Figs 3, 4), dextrotropic and very unequal, a 1st quartette of very small cells 1a-1d is cut off from stem cells 1A-1D. In *Tubifex*, 1a-1d lie bilaterally on either side at the anterior pole, 1d and 1a to the left, 1b and 1c to the right. In *Rhynchelmis*, they are similarly bilateral, but dorsally placed.

The laetotropic 4th division begins with the division of 1D into 2d and 2D (Fig. 6). 2d is much larger than any cell of the 1st quartette, though in *Tubifex* it is only about one-quarter, and in *Rhynchelmis* less than one-quarter, of the size of 2D. In *Tubifex*, 2d pushes forward anterodorsally between 1d and 1c to lie in the dorsal midline, with the 1st quartette cells as an arc around its anterior face. In *Rhynchelmis*, 2d lies mid-dorsally behind the 1st quartette. 2a, 2b and 2c are now cut off laetotropically as small cells, so that 2a is added to the left, 2b to the front and 2c to the right of the micromere arc bordering 2d (Figs 5, 6).

At the same time, the 5th division begins in the D-quadrant. The products of this division as it affects 2D differ in the two embryos. In *Tubifex*, the d component of the 3rd quartette, 3d, is a small cell cut off dextiotropically and added to the left-hand end of the micromere arc (Fig. 5). In *Rhynchelmis*, 3d is cut off in the dorsal midline as a cell directly behind and equal in size to 2d (Fig. 6). The latter cell, on the other hand, behaves similarly in both species at this stage, dividing unequally, with its smaller daughter pushing into the micromere arc while its larger daughter remains mid-dorsal. This is repeated at least twice in subsequent divisions, a cell being added on each occasion to the micromere arc, leaving the large cell in the mid-dorsal line. The final designation of the large cell as $2d^{111}$ in *Tubifex* and $2d^{22}$ in *Rhynchelmis* (Figs 7, 8) does not alter the fundamental similarity of the process in the two species.



Figs 5-8.—5, *Tubifex*, 17-cell stage, dorsal view. 6, *Rhynchelmis*, 12-cell stage, dorsal view. 7, *Tubifex*, 22-cell stage, anterolateral view. 8, *Rhynchelmis*, 30-cell stage, dorsal view. (After Penners, 1922; Swetloff, 1923b.)

Meanwhile, the 5th cleavage division continues in the remaining quadrants, the stem cells 2A, 2B and 2C cutting off the small cells 3a, 3b and 3c to lie on the left, anteriorly and on the right respectively, at the outer rim of the micromere arc (Figs 5, 7). In *Tubifex*, the cells 1c and 1d, on the right and left in this arc, also divide equally, increasing the number of micromeres. In *Rhynchelmis*, they are said not to divide again, but the evidence for this is not positive.

The later part of the 5th division is accompanied by precocious 6th division in the D-quadrant. The stem cell 3D divides into large 4d and 4D cells lying in midline (Figs 7, 8). In *Tubifex*, 4d is equal in size to 4D, although almost all of the yolk in the parent 3D stem cell is confined to 4D. In association with its relatively large size, 4d occupies the posterior midline, pushing 4D ventrally and 3A, 3B and 3C forwards. 3B thus comes to occupy an anterior position, while the micromere arc in front of 2d is lifted into a dorsal position. In

Rhynchelmis, 4d is much smaller than 4D, the two cells being posterodorsal and posteroventral respectively.

In the remaining yolky stem cells, 3A, 3B and 3C, further divisions are equal, simply increasing the number of yolky cells; and 4D now divides in the same way.

After the formation of 4d, the embryos of *Tubifex* and *Rhynchelmis* are remarkably similar in general construction. Each has (Figs 7, 8): (i) ventrally, a number of large yolky cells, the stem cells 3A–3C and 4D, forming a relatively greater proportion of the whole in *Rhynchelmis* than in *Tubifex*; (ii) posterodorsally, one (*Tubifex*) or two (*Rhynchelmis*) large cells in the midline, segregated via 2d in *Tubifex* and via 2d and 3d in *Rhynchelmis*, surrounded anteriorly and laterally by an arc of micromeres cut off from the stem cells as 1st, 2nd and 3rd quartette cells by a similar series of divisions in both species; (iii) posteriorly in the dorsal midline, a large 4d cell, relatively greater in size in *Tubifex* than in *Rhynchelmis*.

The orientation of these cells relative to the major axes is attained in *Tubifex* during cleavage. In polychaetes in which the D cell is disproportionately large, there is a tendency for A and C cells behind the prototroch to be pushed ventrally, and also for the posterior D cell, 4D, to be pushed ventrally as the large 2d and 4d cells are formed, but the anterior part of the embryo in front of the prototroch remains unaffected by this (Anderson, 1966a). In *Tubifex*, these tendencies are exaggerated and in the absence of a prototroch, associated with a large egg in a protective cocoon, they influence even the anterior end. After the formation of the 1st quartette, the axial relations of the eight cells are still as in polychaetes. Formation of a large 2d cell, however, displaces the posterodorsal stem cell 2D posteriorly and ventrally. Formation of a large 4d cell accentuates this, pushing 4D into a ventral position. The B quadrant stem cell is thereby displaced to the anterior end, so that 2b and 3b are cut off upwards at this end, in front of 2d. Similarly, the A and C quadrant stem cells are rotated forwards and upwards, and cut off 2a and 3a, and 2c and 3c, upwards on the left and right of 2d. In this way, the B quadrant becomes anterior, the D quadrant dorsal and posterior, and the A and C quadrants left and right vertical respectively.

It can be suggested that loss of the prototroch is prerequisite to this new pattern of modified spiral cleavage. While the prototroch persists (Polychaeta), modifications associated with an increased size of 2d and 4d posterior to the prototroch have little influence on cells anterior to it. With loss of the prototroch, a new quadrant configuration, established during cleavage in *Tubifex*, becomes possible. In *Rhynchelmis*, in association with a relatively greater yolk mass, the new configuration develops directly.

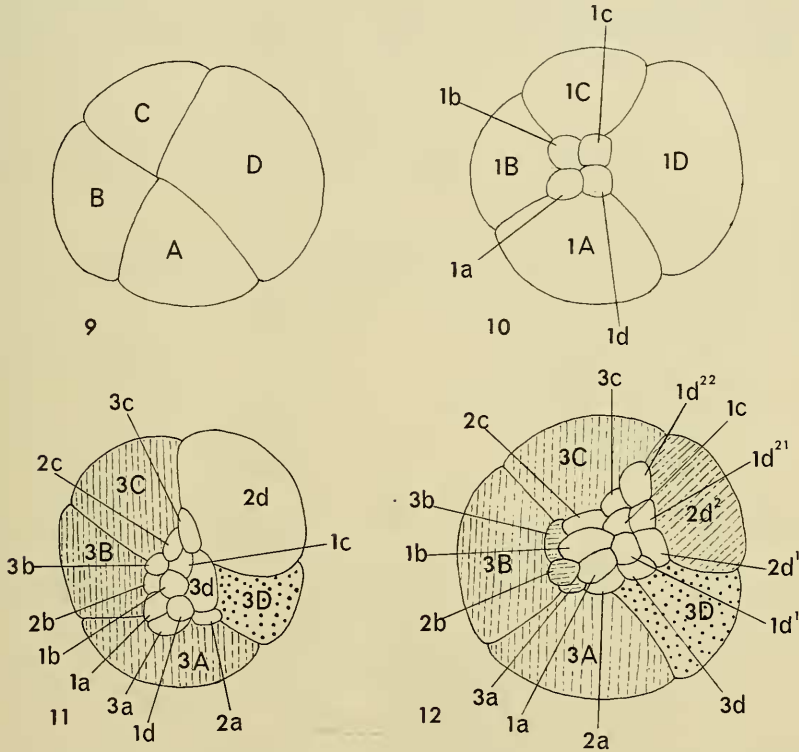
Primitive cleavage in the Hirudinea

From a comparative viewpoint, the important feature of cleavage in primitive leeches is its close similarity in every respect to cleavage in primitive oligochaetes. In *Glossiphonia* (Schliep, 1914) and *Theromyzon* (Schmidt, 1917, 1925a), as in the oligochaete *Rhynchelmis*, the quadrant orientation, B anterior, D posterior, is established after the first two cleavage divisions, and subsequent divisions proceed in a similar manner in the two species. In association with the large size of the egg, 500–1000 μ in diameter in *Glossiphonia complanata*, 600 μ in diameter in *Theromyzon*, the 1st cleavage division is unequal, although less markedly so than in primitive oligochaetes, AB being only a little smaller than CD. At the next division, AB divides equally into anterior B and left-lateral A, while CD divides into right-lateral C, equal in size to A and B, and posterior D, slightly the largest cell (Fig. 9). The parallel with *Rhynchelmis* is unmistakable and continues into subsequent divisions.

The 3rd division is very unequal, a 1st quartette of very small cells being cut off dextrotropically from the stem cells to lie in bilateral arrangement, dorsally

on either side of the midline, 1a and 1d to the left, 1b and 1c to the right (Fig. 10). As in oligochaetes, this division occurs first in the D quadrant, then in the C quadrant, with A and B lagging.

The laeotropic 4th division begins in the D quadrant. 1D divides into a large but yolky-free dorsal cell, 2d, displaced at this stage to the right, and an only slightly larger, but yolky, 2D cell lying posteroventrally and to the left. The three remaining cells of the 2nd quartette are then cut off as micromeres. 2a on the left, 2b anteriorly and 2c on the right, around the 1st quartette, making up a micromere cap (Fig. 11).



Figs 9-12.—9, *Glossiphonia*, 4-cell stage, dorsal view. 10, *Glossiphonia*, 8-cell stage, dorsal view. 11, *Glossiphonia*, 16-cell stage, dorsal view. 12, *Theromyzon*, 19-cell stage, dorsal view. (After Schliep, 1914; Schmidt, 1944.)

In the dextiotropic 5th division (Figs 11, 12), 2D, 2C, and 2A and 2B throw off, in sequence, small 3rd quartette cells 3d, 3c, 3a and 3b, which add to the edge of the micromere cap. 2d also divides unequally, throwing off a small cell 2d¹ on to the posterior edge of the micromere cap, the sister cell 2d² remaining large, posterodorsal and displaced to the right, with 3D lying posteroventrally below it and slightly displaced to the left. At the same time, the 4th division is completed in the 1st quartette, whose cells divide approximately equally into mid-dorsal 1a¹-1d¹ and adjacent 1a²-1d² in the centre of the micromere cap.

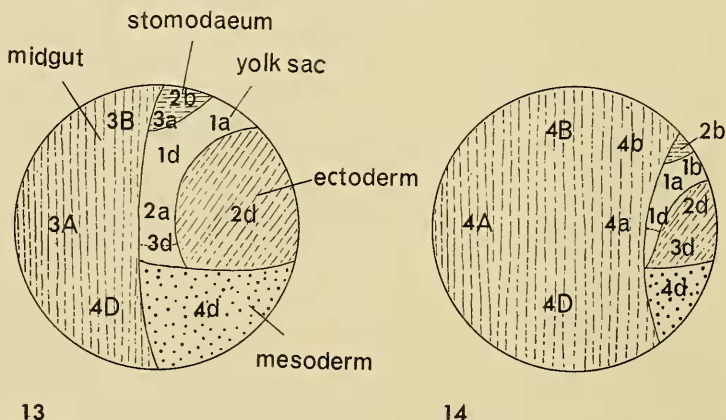
After the formation of 2d² at the 5th division, the embryos of *Glossiphonia* and *Theromyzon* have the same general configuration as those of primitive oligochaetes at a slightly later stage, namely (Fig. 12): (i) ventrally, a number of large yolky cells, 3A-3C; (ii) posterodorsally, a large cell 2d², slightly displaced to the right, with a cap of micromeres anterior to it, cut off from the stem cells as 1st, 2nd and 3rd quartette cells by a similar series of division in both species; (iii) posteroventrally, slightly displaced to the left, a large cell 3D.

The only difference between this pattern and that of oligochaetes summarized in the same way above is the lack of subdivision of 3D in leeches into posterior 4d and ventral 4D. The reason for this difference will be explained below in terms which minimize its significance.

Primitive fate of the cleavage blastomeres in the Clitellata

As in polychaetes (Anderson, 1966a), once the cell 4d has been cut off in oligochaetes from its stem cell, early in the 6th cleavage division, it becomes possible to assign all blastomeres to presumptive areas of specific subsequent fate. In primitive leeches, the cleavage blastomeres can similarly be assigned to presumptive areas by the end of the 5th cleavage division. In both groups, the pattern of presumptive areas is identical. Their most conspicuous feature in comparison with polychaetes is simplification, associated with the absence of presumptive larval organs. The areas comprise (Figs 13, 14, 18): (a) a dorsal area of presumptive ectoderm; (b) at the anterior edge of this area, in the dorsal midline, an area of presumptive stomodaeum; (c) a posterior area of presumptive mesoderm, incorporating presumptive germ cells; (d) a ventral area of presumptive midgut.

In the absence of presumptive prototroch, the separation of presumptive ectoderm into anterior and posterior parts seen in polychaetes is not found in clitellates. Presumptive ectomesoderm is also absent.



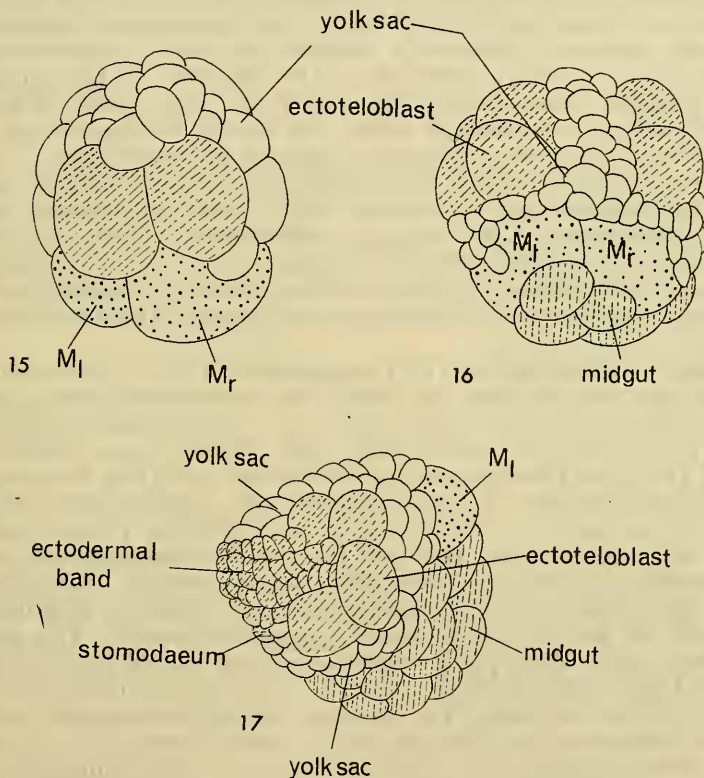
Figs 13-14.—13, presumptive areas of the blastula of *Tubifex*. 14, presumptive areas of the blastula of *Rhynchelmis*. Diagrams based on data from authors listed in the text.

For oligochaetes, the designation of the above areas is supported by the work of Penners (1922, 1924), Penners and Stablein (1930) and Meyer (1929, 1931) on *Tubifex*, Penners (1929) on *Pelosclex benedini*, and Vedjovsky (1888-92), Bergh (1890), Penners (1922), Swetloff (1923b) and Iwanoff (1928) on *Rhynchelmis*. *Pelosclex*, as far as it has been studied, develops in the same manner as *Tubifex*. Differences between *Tubifex* and *Rhynchelmis* occur only in the constitution of the presumptive ectoderm.

Of the four areas listed above, two in oligochaetes correspond in every respect to the homologous areas in polychaetes. The presumptive mesoderm is segregated posteriorly in the single cell 4d which, before it assumes its definitive mesodermal fate, throws off into the interior a pair of small cells, the primordial germ cells. 4d then divides equally bilaterally into M_1 and M_r , teloblasts which subsequently bud off paired ventrolateral mesodermal bands (Figs 15, 16). The presumptive midgut is segregated in the large, ventral, yolky cells 3A-3C and 4D. There also appears to be a close correspondence between the presumptive stomodaeal area of oligochaetes and that of polychaetes. Especially in *Tubifex*, and perhaps also

in *Rhynchelmis*, the anterodorsal micromeres which make up this area stem mainly from 2b and 3b, while 3a can also be implicated in *Tubifex* (Fig. 7). Although the area as a whole is displaced anteriorly, its similarity in relative position and mode of segregation to the stomodaeal presumptive area of polychaetes is conspicuous (compare Anderson, 1966a).

The presumptive ectoderm of oligochaetes is made up of one or, in *Rhynchelmis*, two large, central cells surrounded anteriorly and laterally by an arc of micromeres (Figs 7, 8). In *Tubifex* and *Peloscolex*, the single central cell, the main product of 2d, divides equally bilaterally into a pair of cells, each of which gives rise by further divisions to a transverse row of four ectoteloblasts (Figs 15–17). In *Rhynchelmis*, the anterior central cell, the main product of 2d,



Figs 15–17.—15, *Tubifex*, dorsal view of stage with paired M-cells. 16, *Tubifex*, posterior view of stage during ectoteloblast formation. 17, *Tubifex*, left lateral view of stage after onset of ectoteloblast activity. (After Penners, 1924.)

divides into a pair of cells which migrate laterally, while the adjacent, posterior, central cell, 3d, divides into a pair of cells each of which divides again into three cells in a transverse row with the large 2d cell at the end of it. The eventual product in each case, although resulting from a slightly different series of divisions, is a row of four ectoteloblasts on either side of the midline. Each ectoteloblast now begins to bud off a row of small cells forwards (Fig. 17).

While the ectoteloblasts are forming, the arc of micromeres multiplies and spreads (Figs 15–17): (i) back mid-dorsally, as a strip separating the two ectoteloblast groups; (ii) up towards the mid-dorsal line from its ends, behind the ectoteloblast groups.

The backgrowth mid-dorsally finally meets the upgrowth from the sides. When this stage is reached, the following sub-areas can be distinguished within the presumptive ectoderm: (1) the ectoteloblasts, incorporating presumptive material of the prostomium and cerebral ganglia and the major part of the ectoderm of the peristomium and trunk segments, and already beginning to bud off this material as ectodermal bands; (2) the presumptive temporary yolk sac ectoderm, mid-dorsally and laterally, later incorporated into the segmental epithelium; (3) the presumptive pygidial epithelium, incorporating presumptive proctodaeum, posterior to the teloblasts. This sub-area, as noted above, has a paired bilateral origin and is also of temporary yolk sac function at this stage.

In comparison with the presumptive ectoderm of the polychaetes (Anderson, 1966a, fig. 22), the presumptive mouth region ectoderm and telotroch are absent, the presumptive anterior (prostomial) ectoderm is incorporated into the ectoteloblasts and all extrateloblastic cells have a new presumptive fate as temporary yolk sac cells, although retaining a capacity for forming definitive trunk and pygidial epithelium. The adaptation of the majority of the ectoderm to a temporary yolk sac function is associated with the large volume of presumptive midgut soon to be accommodated within the interior of the embryo. Deletion of the telotroch, as of the prototroch, requires no explanation. Deletion of distinct mouth region ectoderm and incorporation of presumptive prostomium into the ectoteloblasts can be regarded as a simplification made possible by advanced lecithotrophy, based on the ectoteloblast pattern of polychaetes. The presumptive ectodermal pattern of oligochaetes is thus derivable from that of polychaetes, completing the correspondence of all presumptive areas and at the same time revealing a new presumptive area pattern of important comparative value.

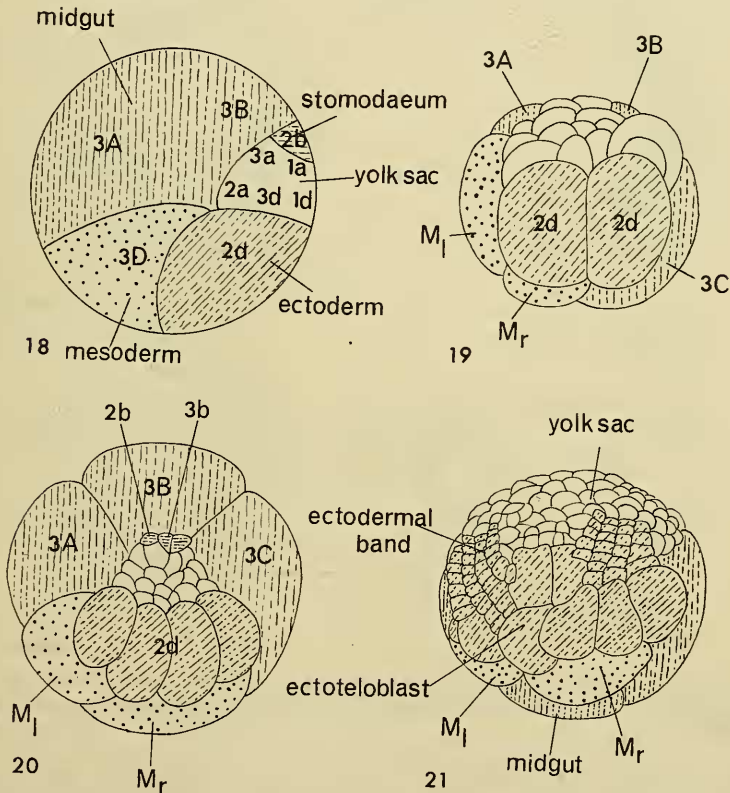
For leeches, while Schliep (1914) and Schmidt (1917, 1925a) provide the data which illustrate the way in which the presumptive area cells become segregated, their assignation to the fates listed above rests with the work of Whitman (1878, 1887), Nusbaum (1884, 1885, 1886), Bürger (1891, 1902), and Bychowsky (1921) on *Glossiphonia* and of Schmidt (1917) on *Theromyzon*, all of whom described aspects of later development. Presumptive mesoderm is segregated precociously in leeches into the cell 3D (Figs 12, 18), but the only significant difference between this and the polychaete-oligochaete condition is that presumptive midgut is eliminated from the D-quadrant. 3D divides equally bilaterally into M_1 and M_r (Figs 19–21), teloblasts which, as in primitive oligochaetes, bud off paired ventrolateral mesodermal bands. The presumptive midgut is segregated, almost as in oligochaetes, into 3A, 3B and 3C (Figs 19, 20) of which the latter cuts off a small ectoderm cell 4c into the micromere cap before assuming its definitive fate. The position of the presumptive stomodaeum (Figs 18, 20) indicates that a major contribution is made to it by the anterior cells of the micromere cap, 2b and 3b, as in *Tubifex*. The presumptive embryonic ectoderm comprises a large dorsal posterior cell and the major part of the micromere cap in front of it. The large cell, as in *Tubifex*, is the main product of 2d and undergoes precocious division in the same sequence, equally bilaterally into four ectoteloblasts on each side (Figs 19–21) which together incorporate presumptive material of the prostomium, cerebral ganglion and the major part of the primordial epithelium of the trunk segments, and begin to bud off eight rows of small cells forwards, four on each side, as ectodermal bands. The micromeres multiply and spread back, between and around the ectoteloblasts and their products (Fig. 21), as in *Tubifex*, forming presumptive temporary yolk sac, later to transform into segmental and pygidial epithelium.

There is thus no doubt that cleavage and presumptive area formation in primitive oligochaetes and primitive leeches are fundamentally identical, supporting the classification of the two together as a single group, Clitellata. They can be interpreted as a particular modification of cleavage and presumptive area formation in polychaetes, associated with enhanced lecithotrophy and direct

embryonic development in a protective cocoon until numerous segments are formed and the adult organization is well established. It will be shown below that it is possible to argue in favour of the derivation of cleavage and presumptive area formation in Onychophora directly from a clitellate-like condition, assuming only further adaptation to increased lecithotrophy. Before pursuing this, however, the stability of presumptive area formation, in spite of secondary egg and cleavage specializations, will be displayed within the clitellates themselves by reference to the earthworms and to gnathobdellid and pharyngobdellid leeches.

Cleavage and presumptive areas in lumbricid oligochaetes

In earthworms, egg size is secondarily reduced, cleavage is modified and little trace of the spiral pattern remains. Attempts to draw comparisons with primitive oligochaetes and with polychaetes by means of the Wilsonian

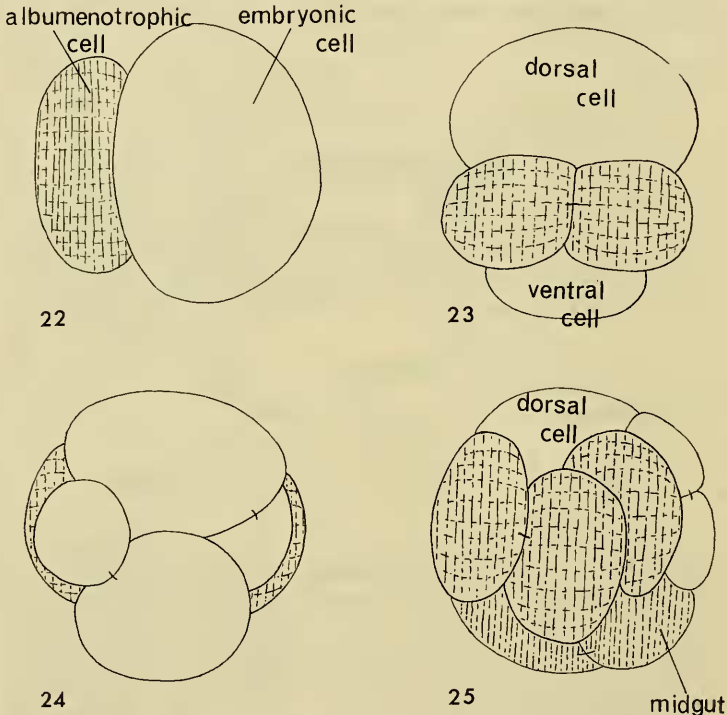


Figs 18-21.—18, presumptive areas of the blastula of *Glossiphonia* and *Theromyzon*. Diagram based on data from authors listed in the text. 19, *Glossiphonia*, postero-dorsal view of stage with paired M-cells. 20, *Theromyzon*, dorsal view of stage during ectoteloblast formation. 21, *Glossiphonia*, postero-dorsal view after onset of ectoteloblast activity. (After Schliep, 1914; Schmidt, 1917, 1925a, 1944.)

enumeration have therefore failed. The results obtained by Swetloff (1923a, 1928) on cleavage in *Bimastus* and *Eisenia*, using this notation, served at the time only to confirm the secondary specialization of earthworm cleavage, the bane of earlier workers (Wilson, 1889; Vedjovsky, 1888-92), without elucidating the mode of this specialization. The same results re-examined today, however, tell a different story, since they show that in earthworms the presumptive areas

of the blastula fall into a pattern identical with that of primitive oligochaetes, although the manner in which they are segregated is new and an additional paedogenetic structure has arisen.

Taking *Bimastus* as an example, cleavage of the microlecithal egg is modified from the very beginning. The 1st division is unequal, yielding a smaller anterior and larger posterior cell (Fig. 22). At the 2nd division, the anterior cell divides equally into right and left daughters, while the posterior cell divides into a larger dorsal and a smaller ventral daughter (Fig. 23). The right anterior cell now divides again, completing a group of three anterior cells which undergo no subsequent division (Fig. 25). These three cells, which are characteristic of earthworms (Hatschek, 1878; Vedjovsky, 1887, 1888-92; Wilson, 1889; Hoffmann, 1899; Swetloff, 1923*a*, 1928) and finally become internal at the



Figs 22-25.—*Bimastus*. 22, 2-cell stage, dorsal view with posterior end on the right. 23, 4-cell stage, anterior view. 24, 7-cell stage, posterior view. 25, 10-cell stage, anterior view. (After Swetloff, 1923*a*.)

anterior end of the embryo before being resorbed, are generally called provisional embryonic excretory cells. They become highly specialized, developing a complex series of intracellular canals, and show pulsatory activity. There is no experimental proof of their excretory function, however, and it is equally possible that they act as absorption and assimilation of albumen until the embryonic gut cells assume this function.

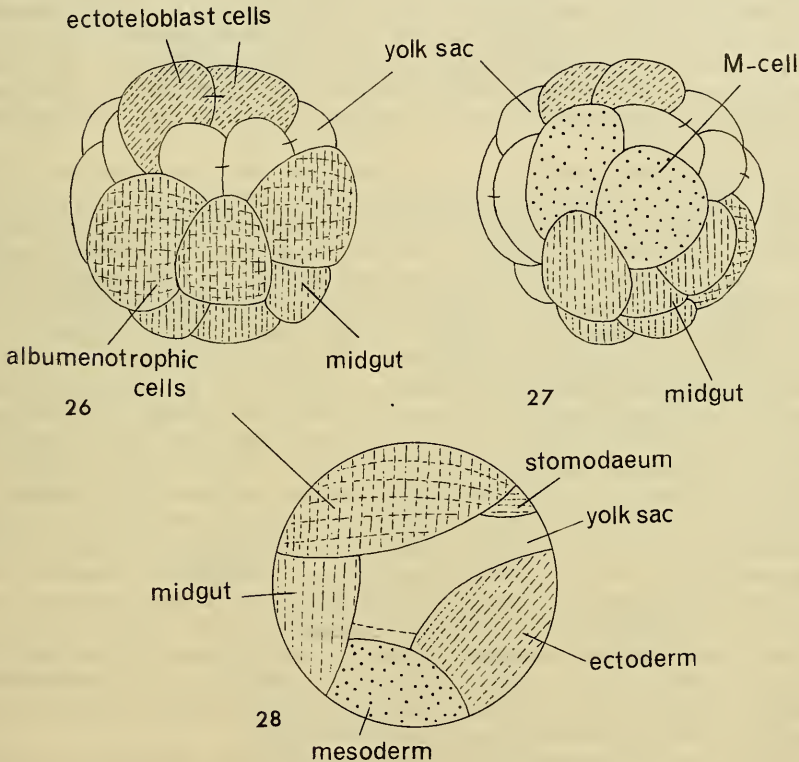
Meanwhile, continuing from the 4-cell stage, the posterior dorsal cell throws off a small cell upwards to the right, while the ventral cell cuts off a small cell upwards to the left (Fig. 24). The latter divides equally to form two small cells on the left, while the ventral cell cuts off a second small cell to the right (Fig. 25).

The large dorsal cell now divides transversely into a smaller anterodorsal and a larger posterior cell. The latter then throws off a small posteroventral cell, somewhat displaced to the right. At the same time, the ventral cell has

divided equally twice, and its products, together with the posteroventral cell, make up a group of five ventral cells containing most of the yolk of the original egg (Fig. 27). These are the presumptive midgut cells. In front of them lie the three excretory cells.

The large posterior cell divides equally bilaterally into two M-cells (Fig. 27), which subsequently bud off ventrolateral mesodermal bands. As in some polychaetes (Anderson, 1966*a*), the M-cells bud off one or two cells added to the presumptive midgut before assuming their definitive fate as mesodermal teloblasts.

The anterodorsal sister of the M-cell mother cell divides equally bilaterally into two cells (Figs 26, 27) which, by further division, give rise to four ectoteloblast cells on each side. The small cells on the right and left multiply and



Figs 26–28.—*Bimastus*. 26, early blastula, anterior view. 27, early blastula, posterior view. 28, presumptive areas of the blastula. (Based on data of Swetloff, 1923*a*.)

spread as a micromere arc in front of the anterodorsal cell (Fig. 26). In this arc, the anterior cells in the mid-line are presumptive stomodaeum, the remainder temporary yolk sac.

When cleavage in *Bimastus* is described in this way, it becomes obvious that the presumptive areas of the blastula (Fig. 28), although segregated by a new series of divisions consequent on secondary yolk loss, are identical in arrangement with those of primitive oligochaetes, the anterior cells having become paedogenetically specialized without disturbing the configuration of the basic pattern. Presumptive midgut is relatively less extensive, commensurate with a reduction in yolk, but is still segregated into a group of cells ventrally. Presumptive mesoderm is contained within a pair of M-cells posteriorly. Presumptive ectoderm dorsally has a large central ectoteloblast mother cell surrounded

anteriorly and laterally by an arc of small cells. Presumptive stomodaeum is confined to the anterior members of this arc, in the midline. The remainder develop as a temporary surface epithelium of the early embryo.

Judging from the position of the excretory cells, lying between presumptive stomodaeum and midgut, it seems likely that they are derived from ancestral presumptive midgut cells, in which case an albumen-absorbing function becomes even more probable.

Cleavage and presumptive areas in gnathobdellid and pharyngobdellid leeches

Like earthworms, the gnathobdellid and pharyngobdellid leeches lay small, microlecithal eggs and show developmental modifications associated with prolonged feeding from an early stage on cocoon albumen (Schmidt, 1944). Cleavage among these forms has been studied only in *Erpobdella atomaria* by Sukatschoff (1903) and Dimpker (1917) and in *Hirudo medicinalis* by Schoumkiné (1953), but the results for the two species are sufficiently alike to suggest a basis of generalization for all gnathobdellids and pharyngobdellids. Not the least interesting aspect of cleavage and presumptive area formation in these animals is its similarity to that of earthworms, evolved in parallel from a fundamentally similar starting point.

In spite of great reduction in egg size, the first three cleavage divisions retain the primitive hirudinean pattern. The 1st division is unequal, giving a larger CD cell and smaller AB cell, the 2nd division unequal in the former, equal in the latter, with the result that a relatively large posterior D cell is flanked by equal A, B and C cells, on the left, anteriorly and on the right respectively. At the 3rd division, a 1st quartette of micromeres is cut off dextiotropically to occupy a dorsal position, bilaterally arranged on either side of the mid-line, 1d and 1a to the left, 1b and 1c to the right (Fig. 29).

Further divisions are now specialized (Fig. 30). The large posterior 1D cell first divides transversely and equally into posterodorsal 2d and postero-ventral 2D cells. The adjacent cell on the right, 1C, cuts off a small cell, 2c, upwards and inwards beneath the 1st quartette. 1A, 1B and 2C now do not divide again. Occupying the anterior and ventral surfaces of the embryo during cleavage, they eventually become overgrown during gastrulation and come to lie internally where they gradually degenerate. These cells clearly recall the three provisional excretory cells of earthworms. Although no mention is made in the work of Sukatschoff, Dimpker or Schoumkiné of any pulsatory activity by the cells, or of the development of intracellular canals within them, it seems likely that their function is similar in leeches and oligochaetes, though whether excretory or absorptive is not yet clear. In leeches, as in earthworms, the relationship of the cells to the other presumptive areas suggests derivation from presumptive midgut, favouring a nutritive function.

From the posteroventral 2D stem cell, a second 3d, and third 4d, cell are cut off into the interior, leaving the large cell 4D still at the surface. Meanwhile, the posterodorsal 2d cell cuts off two small cells 2d¹ and 2d²¹ forwards at the surface behind the 1st quartette, leaving the large cell 2d²² still posterodorsal.

The dorsal micromere cap now begins to multiply and spread (Figs 31, 32). Behind it, the large 2d cell divides equally bilaterally into two, each of which further divides into a typical row of four ectoteloblasts on either side of the dorsal mid-line (Figs 31, 32). Posteroventrally, the large 4D cell also divides equally bilaterally into two, each of which then divides into a lateral cell and a group of three median cells (Fig. 32). The latter push forward into the interior beneath the ectoteloblasts and make contact with the cells previously cut off into the interior from 1C and 2D. The lateral products of the 2D cell are the M-cells, which now begin to bud off mesodermal bands in the usual manner. The internal products of 2D, together with 2c, constitute the presumptive midgut.

Taking into account the above series of divisions and what is known of gastrulation and subsequent development in *Erpobdella* and *Hirudo*, the

group of micromeres. Presumptive stomodaeum is made up by cells at the anterior margin of this group. Furthermore, the parallel with earthworms as a similar response to a similar nutritional situation is unmistakable, and is especially emphasized by the paedogenetic specialization of the "anterior" presumptive midgut cells as provisional embryonic, possibly albumenotrophic cells. In spite of great modification of the cleavage sequence, differently in earthworms and in gnathobdellid and pharyngobdellid leeches, and of changed embryonic nutritional relationships, the basic presumptive areas remain constant in composition, relative juxtaposition and fate.

Cleavage and presumptive areas in other clitellates

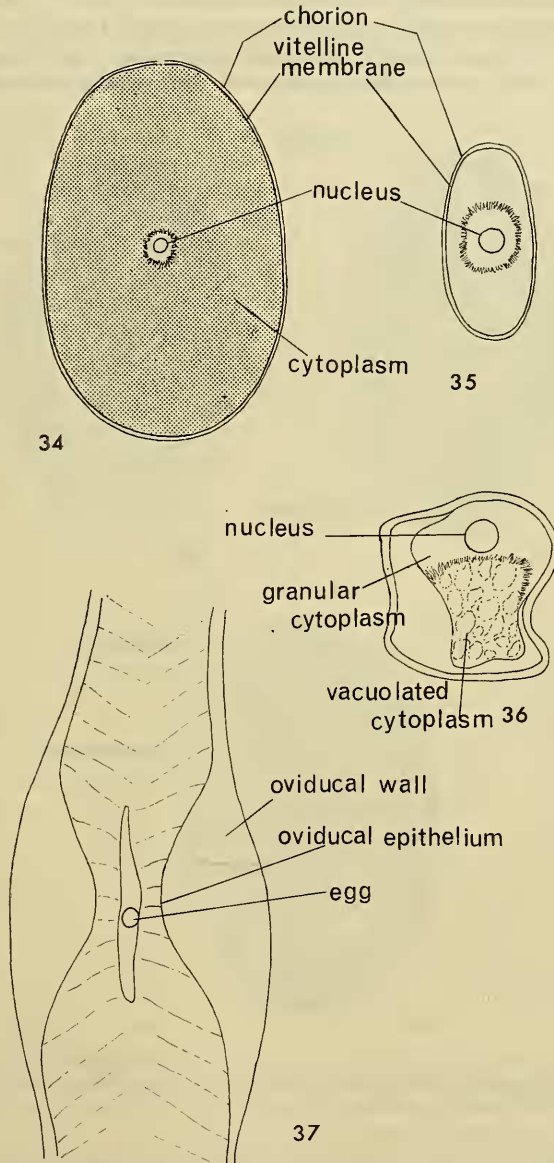
In other clitellates morphologically more primitive than the earthworms and gnathobdellid and pharyngobdellid leeches, namely, the naidid oligochaetes and piscicolid leeches, the eggs are also microlecithal. Here, however, in spite of the systematic position accorded to these families in their respective subclasses, cleavage and the trophic relationships of the embryo in the cocoon are yet more specialized. As the work of Swetloff (1926) and Dawydoff (1942) on naidids and of Schmidt (1921, 1924, 1925*b*, 1930, 1939, 1941, 1944) on piscicolids shows, one of the products of cleavage is a provisional cellular envelope within which ectoteloblasts, M-cells and presumptive midgut become established and express their fate in a normal clitellate manner, and it seems highly probable that the provisional envelope has an absorptive function, rapidly supplemented by further, precociously formed temporary organs subserving feeding on the ambient nutrient albumen of the cocoon. Although it is not intended at the present time to discuss the development of these embryos in detail, there is no doubt that they, too, can be interpreted as modifications of primitive clitellate development in which the basic pattern of presumptive areas is retained in combination with adaptations to albumenotrophy, and they are mentioned here for two reasons. Firstly, they illustrate how even the most aberrant modifications, seemingly totally unintelligible when described by the Wilsonian system of enumeration, can be understood relative to their primitive antecedents when the formation and fate of presumptive areas is recognized. Secondly, like the earthworms and the gnathobdellid and pharyngobdellid leeches, they exemplify the fact that parallel developmental modifications can originate from parallel, related antecedents and need not necessarily be assigned to a common ancestry.

Cleavage in the Onychophora

As has been shown above, cleavage and presumptive area formation in primitive clitellates are adapted to direct lecithotrophic development within a protective cocoon, all known cases of the development of clitellates with small eggs being interpretable as instances of secondary yolk loss, associated with specialized albumenotrophy. Furthermore, lecithotrophic clitellate development manifests a particular modification of the pattern of total spiral cleavage and presumptive area formation seen in yolky polychaete embryos. While it is not possible on this evidence to single out any living polychaete family as a clitellate ancestor, the general notion of a monophyletic relationship between clitellates and polychaetes is supported by the evidence of development. It is also clear that, as in polychaetes, the structure of the presumptive areas making up the wall of the blastula depends on the amount of reserve material incorporated into the egg and that cleavage is modified in ways commensurate with the establishment of these areas.

Now, just as the primitive eggs of clitellate annelids are relatively large and yolky, so too are those of Onychophora. As has been demonstrated especially by Manton (1949), all instances of the development of small eggs in Onychophora reveal evidence of secondary yolk loss. Yolky onychophoran eggs, however, are larger than those of clitellates and individually enclosed in adherent egg membranes, and secondary yolk loss is associated, not with cocoon life, but with viviparity. Concomitantly, the early development of yolky eggs in Onychophora

is more highly modified than that of clitellates, the specializations associated with onychophoran viviparity are different from those associated with clitellate albumenotrophy, and the early embryology of the two groups seems quite dissimilar.

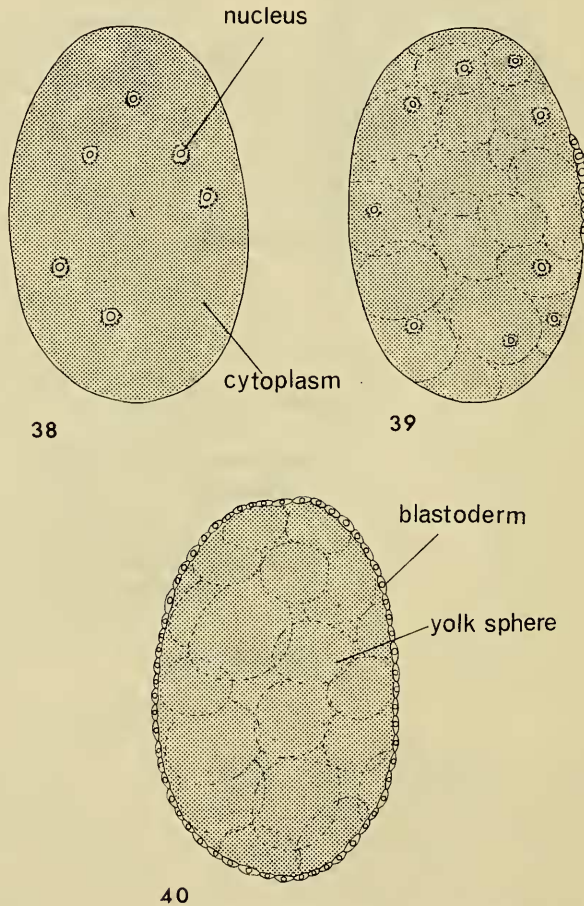


Figs 34–37.—34, diagrammatic longitudinal section through yolk Onychophoran egg. 35, egg of *Peripatopsis balfouri* after dilatation. (After Manton, 1949.) 36, the same egg in transverse section. (After Manton, 1949.) 37, diagrammatic longitudinal section through upper oviduct of placental species, with zygote in lumen.

Only a little is known of the yolk eggs of Onychophora, but it is clear that they combine dimensions of more than 1 mm. (*Ooperipatus*, 1.9 mm., Dendy, 1902; *Peripatoides novae zealandiae*, 1.5 mm., Sheldon, 1888; *Peripatoides orientalis*, 1.3 mm., Anderson, unpublished; *Eoperipatus weldoni*, 1.3 mm.,

Evans, 1902) with a primitive centrolecithal structure, dense with yolk, devoid of periplasm and containing the zygote nucleus in a more or less central cytoplasmic halo (Fig. 34). Two membranes enclose the egg, a thin vitelline membrane and an external chorion which is thick in the oviparous *Ooperipatus*, thin in the ovoviviparous *Peripatoides* and *Eoperipatus*.

The accounts of cleavage given by Sheldon (1888, 1889a) and Evans (1902), although incomplete, show that in yolky onychophoran eggs it follows the typical arthropod mode, with nuclei and associated cytoplasmic haloes dividing and



Figs 38-40.—Yolky onychophoran egg. 38, early cleavage, diagrammatic longitudinal section. 39, emergence of blastomeres at the surface, diagrammatic longitudinal section. 40, blastoderm stage, diagrammatic longitudinal section.

increasing in number in the yolk, then rising to the surface (Figs 38, 39). At the same time, the yolk divides into a number of irregular spheres. Grouped at first as a small disc of blastomeres, the superficial cells spread, with further divisions, to form a blastoderm enclosing the yolk spheres (Fig. 40). According to Sheldon, some of the nuclei and their cytoplasmic haloes in *Peripatoides novae zealandiae* remain within the yolk, but nothing is known of their fate, and Evans (1902) was unable to trace them in *Eoperipatus weldoni*. In general, it appears that cleavage in yolky onychophoran eggs is of a simple centrolecithal type, setting out a blastoderm around a yolk mass.

Development in the secondarily yolkless, viviparous Onychophora follows two modes, non-placental and placental. In the former, described with especial clarity for *Peripatopsis* by Manton (1949), the eggs released from the ovary are small and spherical (*P. sedgewicki*, 65–80 μ ; *P. moseleyi*, 150–172 μ ; *P. balfouri*, 380 μ ; *P. capensis*, 260 μ ; Sheldon, 1889b; Bouvier, 1904; Manton, 1949), but on entry into the oviduct, they swell rapidly and become ellipsoidal (*P. balfouri*, Fig. 35) or cylindrical with hemispherical ends. Their dimensions are then, *P. sedgewicki*, 260 $\times$ 80 μ ; *P. moseleyi*, 520 $\times$ 160 μ ; *P. balfouri*, 480 $\times$ 220 μ ; and *P. capensis*, 600 $\times$ 145 μ (Manton, 1949) and the swelling can be interpreted as a delayed partial recapitulation of the enlargement which occurs in a yolkly onychophoran egg during vitellogenesis. After swelling has taken place, the zygote nucleus lies in an irregular, granular cytoplasmic mass at one side of the egg (Fig. 36). The egg now becomes enclosed in two thin membranes which Manton (1949) suggests are vitelline membrane and chorion.

Early cleavage is similar in all four species (Figs 41–47). The cytoplasm of the egg breaks up into a number of spheres of different sizes, recalling the yolk spheres of *Peripatoides* and *Eoperipatus*, floating freely inside the egg membranes in a watery fluid. The zygote nucleus lies in one of the spheres, while the remainder are anucleate pseudoblastomeres. The onset of cleavage mitoses is thus somewhat delayed. When it occurs, the 1st blastomere divides to form a disc of blastomeres apposed to the inner surface of the egg membranes on one side of the egg, recalling the blastomere disc of the yolkly-egged species. As they form, the blastomeres of the disc gradually absorb the material of the disintegrating pseudoblastomeres. By the time 64 cells are present, the blastomere disc extends as a saddle around two-thirds of the egg circumference, leaving one side and both ends blastomere-free.

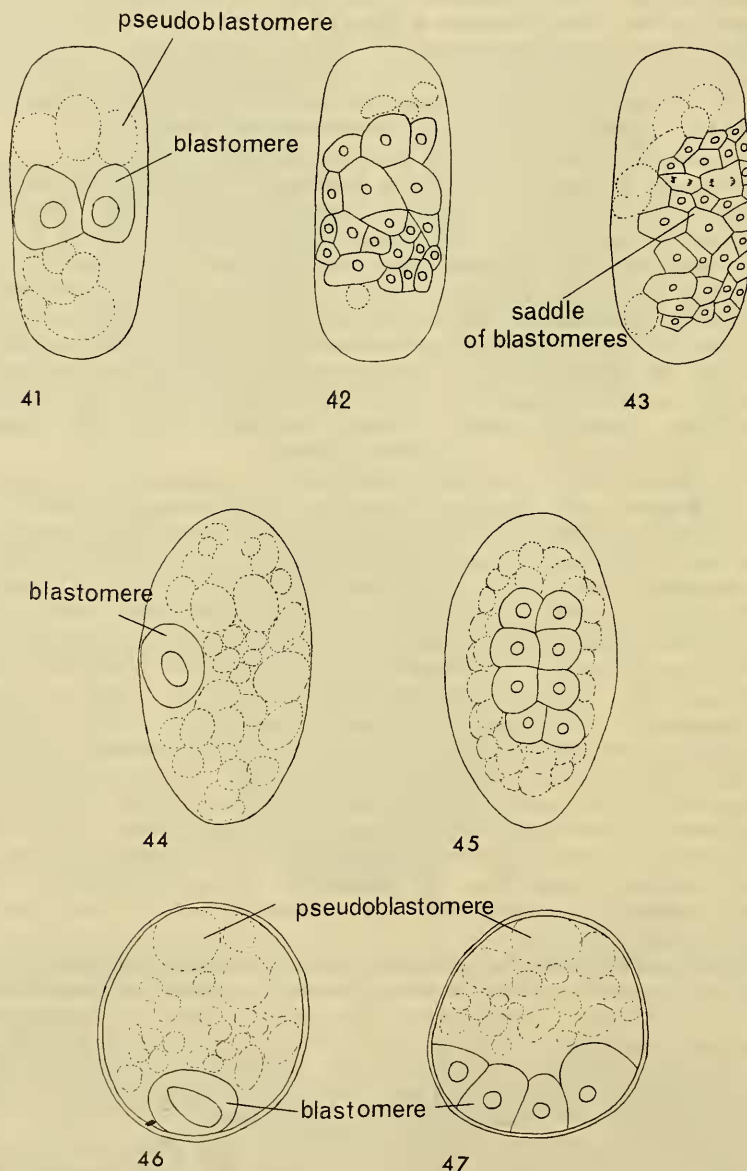
In *P. sedgewicki* and *P. moseleyi*, the saddle of blastomeres now gives rise by further cell division and marginal closure to a blastoderm of about 160 cells around a central fluid-filled space (Fig. 48). Cell division continues and at the same time the vitelline membrane is absorbed and the blastoderm swells by fluid uptake, stretching the outer membrane, until its diameter becomes, in *P. sedgewicki*, about 1000 $\times$ 500 μ , in *P. moseleyi*, about 900 $\times$ 500 μ . The only difference between these embryos and the more primitive yolkly onychophoran embryos at the same stage is replacement of yolk by fluid in the blastocoel.

In *P. capensis*, cleavage and blastoderm formation overlap a precocious onset of movement of cells into the interior (Figs 49, 50). At the edges of the saddle of blastomeres, cells enlarge and move into the concavity of the saddle. This process continues as the saddle cells divide further and spread as a blastoderm, so that the inner surface of the blastoderm becomes lined with large, vacuolated cells. The edges of the outer layer finally form a slit orientated along the anteroposterior axis of the embryo, and with closure of the slit the blastoderm becomes complete. No dilatation of the blastoderm occurs in *P. capensis*, although the surrounding egg membranes swell to the expected size as a result of fluid ingress.

In *P. balfouri* (Figs 51–54), the marginal cells of the saddle of blastomeres also enlarge and move into the interior as the saddle spreads to form a blastoderm, so that the blastocoel is temporarily filled with a mass of vacuolated cells; but these now disintegrate, while at the same time the blastoderm dilates, attaining a final condition like that of *P. sedgewicki* and *P. moseleyi*. The significance of the cleavage peculiarities of *P. capensis* and *P. balfouri* as compared with *P. sedgewicki* and *P. moseleyi* will be taken up below.

The placental viviparous Onychophora have eggs which are even smaller than those of the non-placental species and, furthermore, lack the recapitulatory swelling of the egg after release from the ovary (Fig. 37). Dimensions lie between 25 μ and 40 μ (Bouvier, 1904) and enclosing membranes are absent (Kennel, 1884, 1885; Slater, 1888). Cleavage in these eggs is little understood. Kennel (1884, 1886) and Slater (1888) showed, and further unpublished observations

by Manton and myself have confirmed, that it is total and equal, resulting in the formation of a morula lying in the lumen of the oviduct (Fig. 55). With further cell divisions, the morula becomes hollow, with a wall one cell thick, before attaching to the oviducal wall (Fig. 56). Rather than dilating to form a blastoderm, the hollow vesicle enters immediately into a further specialized phase, in which the cells in the region of attachment multiply to form a hollow stalk addressed terminally as a flat placental plate against the oviducal wall,



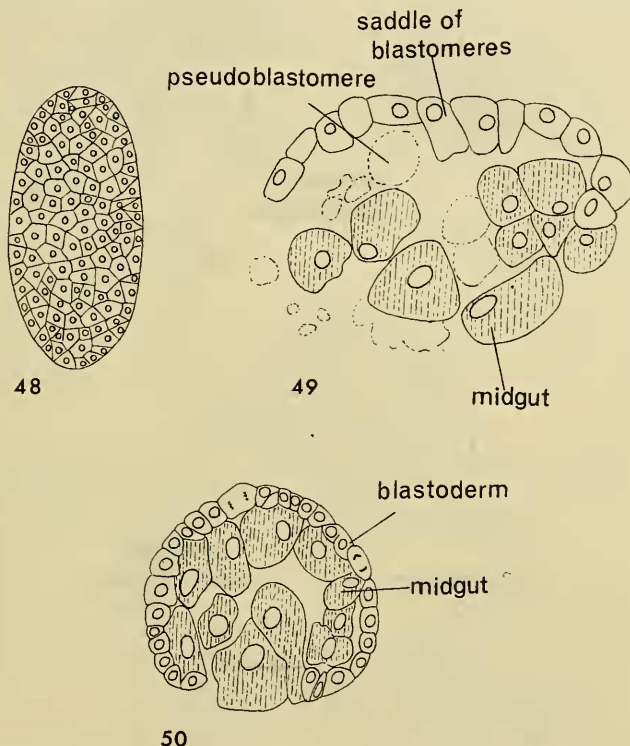
Figs 41-43.—*Peripatopsis moseleyi*. 41, 2-cell stage. 42, 21-cell stage. 43, saddle of blastomeres (After Manton, 1949.)

Figs 44-47.—*Peripatopsis balfouri*. 44, 1-cell stage. 45, 8-cell stage. 46, 1-cell stage, transverse section. 47, 16-cell stage, transverse section. (After Manton, 1949.)

while the remaining cells multiply to increase the size and cell number of the hollow vesicle at the free end of the stalk (Fig. 57). The further development and comparative significance of the stalk and vesicle will be examined below.

Developmental fate of the cleavage blastomeres in Onychophora

Just as in the Clitellata, the cells of the blastoderm or its equivalent immediate pre-gastrula stage in Onychophora can be assigned to presumptive areas of specific subsequent fate by taking account of the events of gastrulation and later development. The focal point in the elucidation of these presumptive areas is the composition of the blastoderm in yolky onychophoran embryos. Unfortunately, one of the weaknesses of Sheldon's work on *Peripatoides novae*

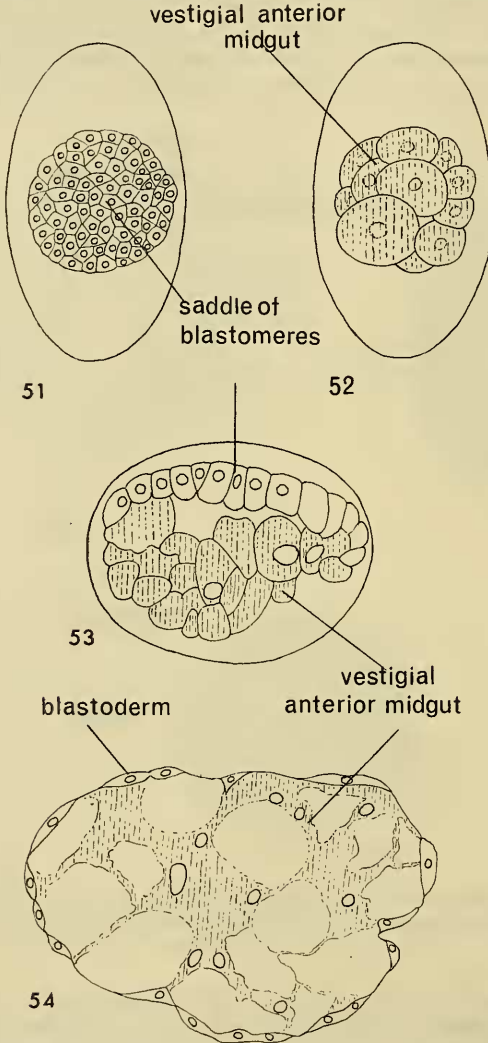


Figs 48-50.—48, *Peripatopsis moseleyi*, blastoderm stage before dilatation. 49, *P. capensis*, section through late cleavage stage. 50, *P. capensis*, section through almost completed blastoderm. (After Manton, 1949.)

zealandiae, and a matter calling urgently for reinvestigation, is a lack of accurate description of gastrulation and early segment formation. At the same time, Sheldon's results, together with those of Evans (1902) and some preliminary unpublished observations of my own on embryos of *Peripatoides orientalis* from New South Wales, permit the tentative construction of a presumptive area map for ovoviviparous species (Fig. 58). Anteroposterior and dorsoventral axes are fixed, the former corresponding to the long axis of the egg. Midventrally lies a long narrow band of presumptive midgut cells, soon to migrate into the interior, leaving an elongate slit at the surface. At the margin of the presumptive midgut lie, anteriorly, presumptive stomodaeum, laterally, paired ventrolateral bands of temporary yolk sac ectoderm and, posteriorly, presumptive proctodaeum. Midventrally behind the latter lie small areas of presumptive posterior midgut and presumptive mesoderm. Associated with the latter are presumptive germ

cells. Lateral to the ventral areas listed above, and converging posteriorly on the presumptive mesoderm, are two broad bands of presumptive ectoderm. Dorsally lies a further broad area of temporary yolk sac ectoderm.

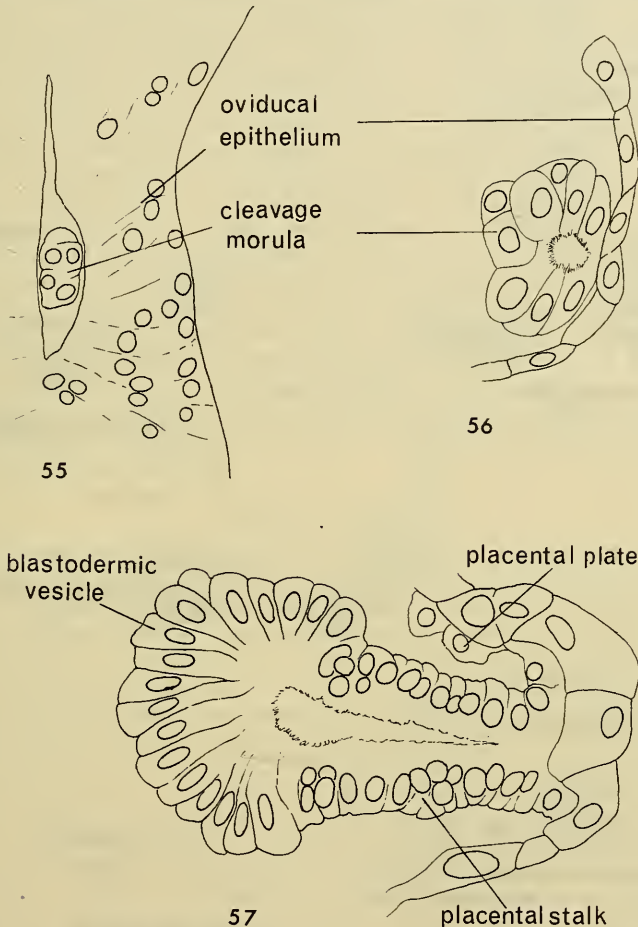
The data of Manton (1949) permit much more firmly substantiated presumptive area maps to be drawn for the viviparous non-placental *Peripatopsis*. In no case in this genus does the anteroposterior axis of the embryo correspond



Figs 51-54.—*Peripatopsis balfouri*. 51, saddle of blastomeres stage. 52, same embryo from opposite side, showing vacuolated cells. 53, transverse section through early saddle of blastomeres stage. 54, transverse section through blastoderm stage. (After Manton, 1949.)

to the long axis of the egg. It may fall in any direction, suggesting that the axial relations of the embryos are established late, after blastoderm formation, by processes which are at present obscure. This, however, does not confound the reality of the presumptive areas which can be discerned. In *P. sedgewicki* they centre on a terminal, and in *P. moseleyi* a lateral cell group which lie postero-ventrally relative to future development. The cells of the group (Fig. 59)

comprise presumptive mesoderm, presumptive posterior midgut anterior and internal to the mesoderm, presumptive ectoderm marginally, and a number of presumptive germ cells. The latter tend to lie between the presumptive midgut and presumptive mesoderm in *P. moseleyi* and at the posterior end of the presumptive mesoderm in *P. sedgewicki*, a minor discrepancy of localization with no special significance. Immediately anterior to the presumptive posterior midgut lie the presumptive proctodaeum and stomodaeum, conjoined. Lateral to them are ventrolateral bands of presumptive temporary yolk sac ectoderm, followed by

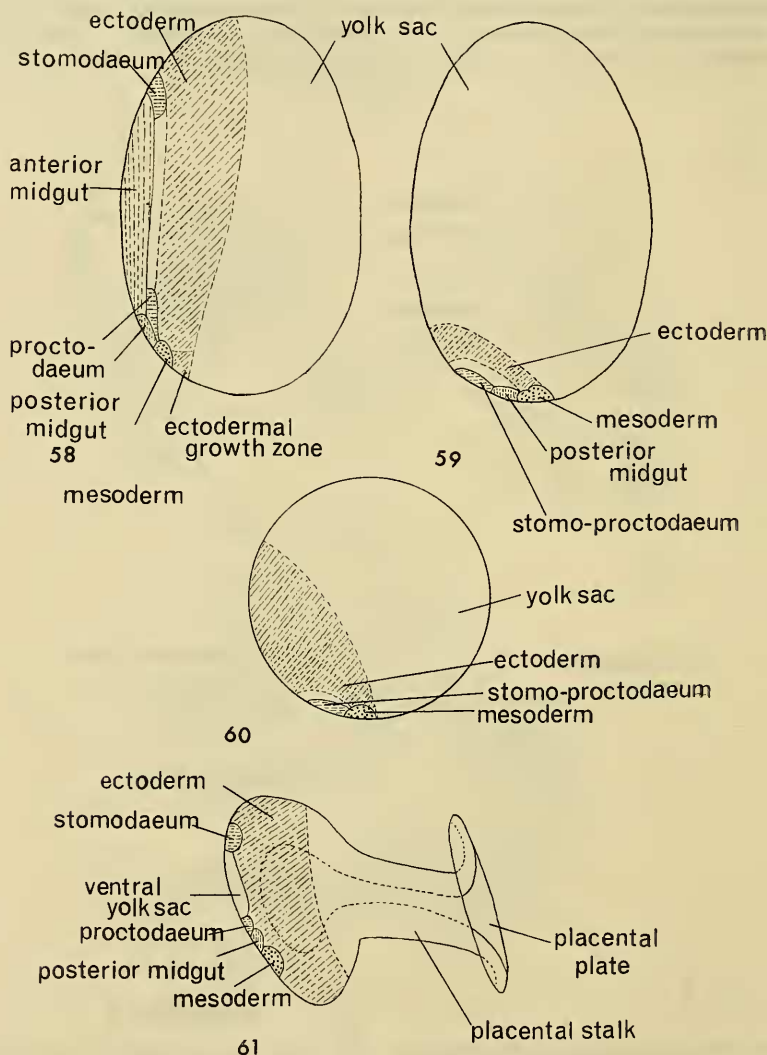


Figs 55-57.—Viviparous placental onychophoran. 55, early cleavage morula. 56, later morula attached to oviducal wall. 57, blastodermic vesicle and placental stalk.

broad bands of presumptive embryonic ectoderm, with the remainder of the blastoderm, laterally and dorsally, constituting further temporary yolk sac ectoderm.

The blastodermal presumptive areas of *P. balfouri* are identical with those described above, with the presumptive germ cells arising from the presumptive mesoderm as in *P. sedgewicki*. It is necessary to take into account, however, the temporary vacuolated cells budded off in *P. balfouri* from the edge of the saddle of blastomeres into the interior during blastoderm formation. Since the point towards which these proliferating edges converge and close subsequently forms the presumptive stomodaeum and proctodaeum, a place can be assigned

along the ventral midline of the latter as the site of origin of the temporary vacuolated cells, already budded off. By comparison with the presumptive area map for yolkly species, the temporary vacuolated cells would appear to be modified midgut cells, as suggested by Manton (1949).



Figs 58-61.—58, presumptive areas of the blastoderm of yolkly Onychophora, diagrammatic lateral view. 59, presumptive areas of the blastoderm of *Peripatopsis sedgewicki*, diagrammatic lateral view. 60, presumptive areas of the blastoderm of *P. capensis*, diagrammatic lateral view. 61, presumptive areas of the blastoderm of viviparous placental Onychophora, diagrammatic lateral view. (Figs 59 and 60 based on Manton, 1949.)

In *P. capensis*, where the blastoderm remains undilated, its presumptive areas show some differences from the preceding species (Fig. 60). Presumptive mesoderm, with which the presumptive germ cells are again associated, remains the same, as do the presumptive proctodaeum and stomodaeum in front of the mesoderm, ventrolateral temporary yolk sac on either side, embryonic ectoderm as paired lateral bands and further yolk sac ectoderm dorsally. The

yolk sac component, however, is not attenuated, and a presumptive posterior midgut area is absent. The midgut rudiment has already become internal, since it is formed by the cells which migrate into the interior from the edges of the saddle of blastomeres during cleavage and line the blastoderm internally. Midgut formation in this way, like the temporary midgut formation of *P. balfouri*, can be interpreted as a precocious budding from the future midline of the presumptive stomodaeum and proctodaeum, the point at which the blastodermal edges converge and fuse.

For the viviparous placental Onychophora, the work of Kennel (1884, 1885) and Slater (1888) and a few unpublished personal observations lead to the following comments on presumptive areas (Fig. 61). By the time that these areas become discernible, the dorsal temporary yolk sac ectoderm has already grown out as placental stalk and plate, as well as occupying the dorsal surface of the blastodermic vesicle. The remaining areas show the expected orientation, but are small and compressed together. Presumptive mesoderm lies postero-ventrally, with presumptive posterior midgut in front of it, then presumptive proctodaeum. Presumptive ventral temporary yolk sac ectoderm occupies the ventral midline, with presumptive stomodaeum anterior to it. Laterally and anteriorly lie paired bands of presumptive ectoderm. The similarity of this presumptive area pattern to that of other Onychophora is clear, in spite of great differences in egg size and mode of cleavage, and is analogous to the similarities described above for the presumptive areas of yolky and secondarily yolkless clitellate embryos. In fact, throughout the Onychophora, in spite of great variations in mode of cleavage, the configuration of presumptive areas shows the same stability as in clitellates, only the presumptive midgut and temporary yolk sac ectoderm, the two areas intimately involved with the yolk, being much affected by secondary yolk loss.

GASTRULATION

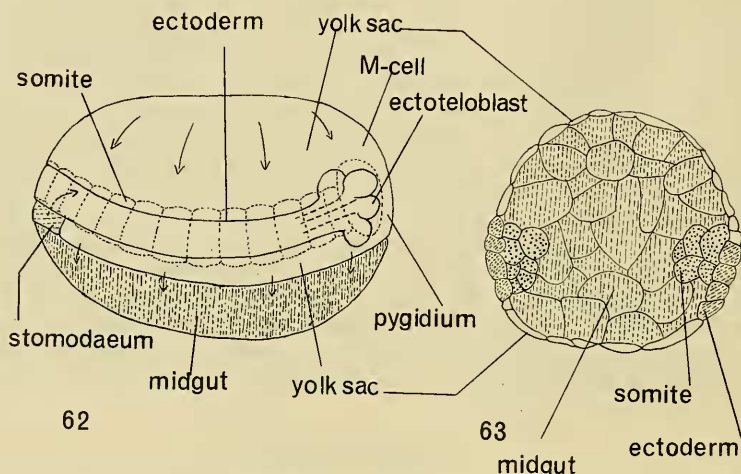
The considerations which have been presented so far permit the recognition of basic presumptive area patterns for the clitellate blastula and the onychophoran blastoderm. Comparison of the two patterns reveals, in the first instance, little in common between them. Presumptive areas, however, have not only a location but a fate, the expression of which begins with gastrulation. Indeed, gastrulation is most satisfactorily defined as the migration of the presumptive areas of the blastula or blastoderm into their organ-forming positions, consequent upon which the fate of the cells of each area is expressed in organ formation itself. This, of course, is not a new definition, being the one which has been employed by workers on vertebrate embryos for many years, but its applicability to invertebrate embryos has only recently been recognized (Anderson, 1962, 1966*a*, 1966*b*). When the parameters, not only of location and fate, but also of mode of expression of this fate, are brought into consideration, the seeming dissimilarity between primitive clitellate and primitive onychophoran embryos all but vanishes. The gastrulation phenomena of the albumenotrophic clitellate embryos, through which a microlecithal blastula becomes transformed into a precocious feeding stage with temporary functional organs, although they can be interpreted as secondary modifications of the gastrulation sequence of primitive lecithotrophic clitellate embryos, will not be discussed in the present context, since they are not pertinent to the comparison with Onychophora.

Primitive gastrulation in the Clitellata

Commensurate with the similarity of their presumptive areas, gastrulation takes an identical course in primitive oligochaetes and primitive leeches. Accounts of gastrulation have been given for *Tubifex* by Penners (1924) and Meyer (1929), for *Peloscolex* by Penners (1929) and for *Rhynchelmis* by Kowalevsky (1871), Vedjovsky (1888-92) and Iwanoff (1928). The work of Whitman (1878, 1887) on *Glossiphonia* and Schmidt (1917) on *Theromyzon*, together with some fragments

in the papers of Nusbaum (1886) and Bürger (1902), provides corresponding information in the glossiphoniid leeches. Together, these studies yield a reasonably detailed picture, though further investigation is desirable.

As might be expected (Figs 62, 63), the large, yolk presumptive midgut rudiment becomes internal as a result of overgrowth by other cells. Cell divisions continue while the rudiment is still exposed at the surface, producing a solid mass of polygonal yolk cells. Presumptive ectoderm, as it continues to develop, spreads down on either side of this mass, eventually meeting in the midline ventrally to enclose it. Complete enclosure is not achieved until most of the segments formed in the embryo have been delineated.



Figs 62-63.—62, gastrulation in yolk-clitellate embryo, diagrammatic lateral view. 63, transverse section through gastrula of *Tubifex*. (After Kowalevsky, 1871; Whitman, 1878; Penners, 1924; Meyer, 1929.)

The M-cells, as in yolk polychaetes (Anderson, 1966a), begin their teloblastic activity while still at the surface, so that budding overlaps the gastrulation movements of these cells into the interior. The latter comes about partly by an inward sinking of the M-cells on either side of the midgut posteriorly, but mainly by overgrowth by the posterior edge of the presumptive ectoderm, which spreads back over the M-cells as far as the edge of the exposed presumptive midgut.

The presumptive stomodaeum, formed as a flat plate of cells at the surface in front of the presumptive midgut, grows into the interior by cell division as a solid mass, formation of a lumen being much delayed.

The overgrowth and enclosure role of the presumptive ectoderm in primitive clitellate gastrulation is attained mainly through cell division and attenuation of the temporary yolk sac component of this area. The temporary assumption of this form by the majority of the presumptive ectoderm is, in fact, a primitive adaptation to rapid completion of an ectodermal surface layer around a large mass of yolk midgut. The ectoteloblasts, in contrast, proceed directly into processes underlying adult organogeny, while being moved from a dorsolateral to a lateral position as the temporary yolk sac ectoderm spreads. As already indicated, they begin their teloblastic activity as soon as they are formed, budding off four parallel rows of cuboidal cells on either side which push forwards towards the anterior end. The cell rows remain superficial, cleaving a path through the temporary yolk sac ectoderm, which at the same time pushes them ventrally as it spreads. The ectoblast rows of each side eventually meet in the midline, first anteriorly in front of the stomodaeal rudiment, then midventrally behind

the stomodaeum and progressively more posteriorly, completing enclosure of the midgut. Most of the temporary yolk sac ectoderm now lies dorsally between the two ectoblast bands, but a narrow strip also persists along the ventral edge of each band and, at the completion of gastrulation by midventral ectodermal fusion, comes to occupy the ventral midline as far forward as the stomodaeal rudiment. Posteriorly it remains contiguous with the pygidial temporary yolk sac ectoderm.

As the ectoblast bands grow in length, the mesodermal bands budded from the M-cells grow beneath them and are similarly carried, first to a lateral, then to a ventrolateral position. The development of these bands as paired segmental somites proceeds in anteroposterior succession and precedes corresponding segment delineation of the overlying ectoblast bands. Later in development, the temporary yolk sac ectoderm is incorporated segmentally into the segmental epithelium, save for the most posterior part, which transforms into the pygidial epithelium. A short intucking of the latter at the anus gives rise to a rudimentary proctodaeum.

Gastrulation in the Onychophora

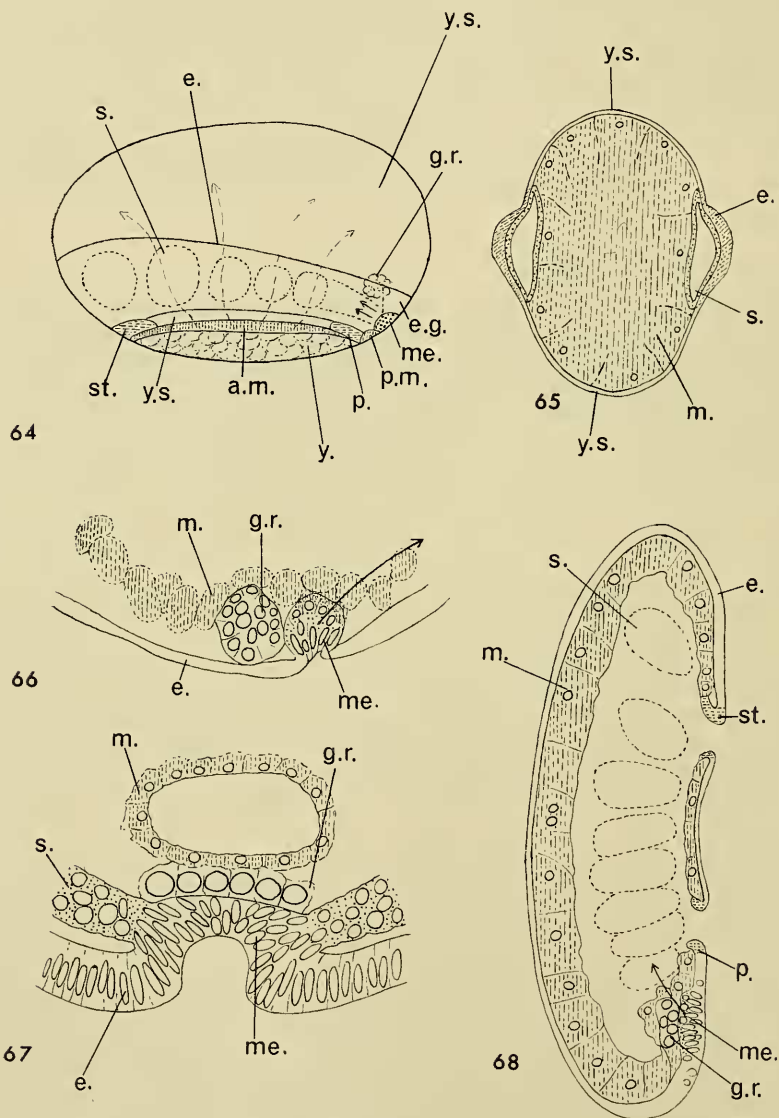
Unlike the clitellates, whose basic gastrulation sequence is adequately displayed by reference to lecithotrophic examples alone, the Onychophora call at the present time for consideration both of yolky and of secondarily yolkless species. As pointed out above, only the work of Manton (1949) on the secondarily yolkless *Peripatopsis* provides modern histological data on the early embryology of the group, so that to approach onychophoran gastrulation without reference to this work would be to ignore almost all of the pertinent evidence. Fortunately, as will be shown below, onychophoran gastrulation is little modified in consequence of secondary yolk loss, so that the events in *Peripatopsis* have an immediate bearing on primitive onychophoran gastrulation and its comparison with clitellates.

In yolky onychophoran embryos, where all presumptive areas are composed of small cells at the surface of a yolk mass, the direct entry of whole areas into the interior, conforming to the definition of gastrulation given above, is at a minimum. For the most part, cells which become internal are budded off from presumptive areas which retain a more or less superficial position and enter directly into organogeny. This condition is also retained in the secondarily yolkless species, the only exceptions being a precocious gastrulation activity of the presumptive midgut in *Peripatopsis capensis* and its vestigial equivalent in *Peripatopsis balfouri* (see below).

The presumptive midventral midgut in yolky embryos seems to retain a definite gastrulation movement (Fig. 64), sinking inwards at the onset of gastrulation, then separating to either side leaving a long midventral slit through which the yolk is exposed at the surface (Sheldon, 1888, 1889a; Evans, 1902; unpublished personal observations). The paired bands of sunken presumptive midgut cells become mitotically active, budding off further cells which migrate outwards and upwards through the peripheral yolk, acting temporarily as vitellophages and becoming enlarged and diffuse as they move, but eventually enclosing the yolk mass and forming the anterior part of the definitive midgut epithelium. At the same time, the presumptive posterior midgut area sinks slightly inwards and begins to proliferate cells which add to the midgut epithelium at the posterior end of the yolk (Fig. 64). It is likely that the latter activity is prolonged, resulting in progressive extension of the midgut as the trunk segments develop, but is completed before the full number of trunk segments is formed.

In *Peripatopsis sedgewicki*, *P. moseleyi* and *P. balfouri* (Manton, 1949), cells proliferated from the slightly invaginated presumptive posterior midgut form the entire midgut epithelium, spreading forwards and upwards beneath the

blastoderm to enclose a central fluid-filled space, and continuing to be proliferated until almost all of the trunk segments are formed (Fig. 68). The mid-ventral temporary vitellophage component of the midgut is lost in *P. sedgewicki* and *P. moseleyi* and only vestigially retained, with precocious proliferation during cleavage, in *P. balfouri*. In *Peripatopsis capensis*, in contrast, the mid-ventral component comes to line the inner surface of the blastoderm during cleavage,



Figs 64-68.—64, gastrulation in yolkly onychophoran embryo, diagrammatic lateral view. 65, transverse section through yolkly onychophoran gastrula. 66, sagittal section through proliferating presumptive mesoderm of *Peripatopsis moseleyi*. 67, transverse section through proliferating presumptive mesoderm of viviparous placental onychophoran. 68, sagittal section through embryo of *Peripatopsis balfouri*. (a.m., anterior midgut; e., ectoderm; e.g., ectodermal growth zone; g.r., genital rudiment; m., midgut; me., presumptive mesoderm; p., proctodaeum; p.m., posterior midgut; s., somite mesoderm; st, stomodaeum; y, yolk; y.s., temporary yolk sac ectoderm.) (Figs 66 and 68 after Manton, 1949.)

as described above, and develops directly as midgut epithelium, the posterior midgut area being lost.

The midgut in viviparous placental species again arises by prolonged proliferation from the presumptive posterior midgut area (Kennel, 1884, 1885; Selater, 1888; unpublished personal observations). After a slight insinking at the onset of gastrulation, the area proliferates cells which spread forwards and upwards to form an elongated epithelial sac. No trace of a midventral component is retained in these species.

In respect of onychophoran midgut development, then, gastrulation in the sense of movements of presumptive areas is slight and each one progresses quickly into proliferation as a precocious onset of organogeny. The significance of this as an adaptation to lecithotrophy will be discussed below.

The same feature characterizes entry of mesoderm into the interior in Onychophora and in the case is accompanied by consistent behaviour in all species (Sedgewick, 1885–1888; Sheldon, 1888; Kennel, 1884, 1885; Evans, 1902; Manton, 1949). The presumptive mesoderm invaginates a little and commences active proliferation, budding off two streams of cells which migrate forwards along bilaterally symmetrical ventrolateral paths (Figs 64–68). Proliferation continues until the mesoderm of all segments is established, by which time of course the anterior parts of the mesodermal bands are well advanced into organogeny. The unmodified persistence in secondarily yolkless embryos of a mode of mesoderm development obviously adapted to extreme lecithotrophy is in marked contrast to the variations in midgut development. Presumptive germ cells also show consistent gastrulation activity throughout the Onychophora, in spite of minor differences of localization in different species. Early in gastrulation they migrate inwards to lie immediately internal to the area of proliferating mesoderm (Figs 66, 67, 68), both in yolky and in secondarily yolkless species (Manton, 1949; unpublished personal observations). Once again, the basic adaptation to lecithotrophy persists with loss of yolk. In *Peripatopsis moseleyi*, where the cells are initially associated with presumptive posterior midgut, they become distinct before the midgut area begins to proliferate, moving into the interior and differentiating by nuclear enlargement, development of nucleoli and reduction to cytoplasmic basophilia, a characteristic germ cell pattern. In *P. sedgewicki*, *P. balfouri* and *P. capensis*, where the initial association is with presumptive mesoderm, differentiation of the germ cells follows migration into the interior as part of the first products of the mesodermal area, but subsequently proceeds in the same way.

Gastrulation activity in the stomodaeal and proctodaeal presumptive areas is closely linked with that of the presumptive midgut. In contrast to clitellate primitive gastrulation, in which the stomodaeal and proctodaeal areas remain superficial and penetrate into the interior by growth and cell division before developing lumina, these areas in yolky onychophoran embryos can be surmised, from the work of Sheldon (1888, 1889a) and Evans (1902), to invaginate shallowly at the ends of the midventral presumptive midgut invagination (Fig. 64). The lateral lips of this invagination separate from the edges of the midgut rudiment as these come together in the ventral midline below the yolk, and unite midventrally at the surface to establish the broad ventral band of temporary yolk sac ectoderm (Fig. 65). At each end, the stomodaeal and proctodaeal invaginations are thereby closed off as short tubes, each of which now proceeds to elongate and push inwards by cell division.

The results of Manton (1949) can be interpreted to show that in *Peripatopsis*, in spite of the absence of a midventral presumptive midgut component in the blastoderm, much of this sequence is retained. The confluent stomodaeal and proctodaeal presumptive areas invaginate together, producing a short midventral slit. The slit elongates (just how it does this is not clear, but ventral temporary yolk sac ectoderm cells appear to multiply to form its lateral lips) pushing the

stomodaeal and proctodaeal rudiments apart. At the same time, the midgut cells below the surface separate along the midventral line and establish contiguity with the edges of the slit so that the latter opens into the midgut cavity. The similarity of this to the corresponding stage described by Sheldon and Evans in yolky embryos is remarkable, although it is attained in a different way in association with presumptive midgut modifications related to secondary yolklessness. Further steps proceed as in yolky species. The midgut epithelium separates once again from the lateral lips of the ventral slit and closes midventrally. The lips themselves also close midventrally at the surface, establishing the narrow midventral band of temporary yolk sac ectoderm found in these species and completing the tubulation of the short stomodaeum and proctodaeum (Fig. 68). Elongation of the latter by cell division now proceeds.

In viviparous placental Onychophora, in association with extreme secondary reduction of the blastoderm, stomodaeal and proctodaeal gastrulation activity is greatly simplified, each area invaginating directly as a short tube (Kennel, 1884, 1885; unpublished personal observations).

Presumptive embryonic ectoderm in Onychophora, formed in situ in the blastoderm as paired ventrolateral ectodermal bands (Figs 64, 65), shows no gastrulation activity. The dorsal component of the temporary yolk sac ectoderm also lacks this activity. The paired ventral component in yolky and secondarily yolkless non-placental species comes together in the ventral midline during gastrulation as described above, to complete the ectodermal covering of the embryo. In placental species, this component is narrow and unpaired and forms directly in its definitive position. In general, the presumptive ectoderm in Onychophora proceeds directly into organogeny after formation, through blastoderm formation, as an almost complete covering epithelium. The spread of ectoderm in gastrulation following total spiral cleavage, as in clitellates, is obviated by this adaptation.

Comparison of gastrulation activities in the three main types of embryo in Onychophora only serves to emphasize, as in annelids (Anderson, 1966a and above), the remarkable stability of their presumptive areas irrespective of great differences in egg size, mode of embryonic nutrition and associated modifications of cleavage. The presumptive areas of all species are not only set out in the same relative juxtaposition but also proceed into the same lines of developmental activity; and where they do not, the differences are easily explained. Presumptive embryonic ectoderm, mesoderm and germ cells develop consistently in all species, irrespective of yolk loss. The dorsal component of the temporary yolk sac ectoderm may remain unexpanded following yolk loss (*Peripatopsis capensis*), but is always retained. The paired ventral component, in spite of yolk loss, maintains its role in assisting stomodaeal and proctodaeal invagination, except in the very small embryos of placental species, where it is greatly reduced. Stomodaeum and proctodaeum formation proceed in viviparous non-placental species in the same manner as in yolky species, and in a manner in placental species which is a direct simplification of this. The presumptive midgut is more affected by yolk loss, but only in simple ways. In most cases, the midventral area, which in yolky embryos produces the cells acting temporarily as vitellophages, is absent (retained vestigially in *Peripatopsis balfouri*) and midgut formation devolves wholly upon the posterior proliferative area. In *Peripatopsis capensis* the reverse has occurred, the posterior area being absent. Both types of modification are simplifications of the primitive condition of a dual development of midgut cells subserving temporary vitellophage function in a large yolk mass anteriorly and simultaneous elongation of the gut posteriorly, and both are equally advantageous in the absence of yolk.

In short, young onychophoran embryos are much less different from one another than they seem and the developmental peculiarities of the various types of yolkless embryo can be interpreted as simple derivations of a basic yolky type.

THE TRANSITION FROM SPIRAL TO CENTROLECITHAL CLEAVAGE AND
THE ORIGIN OF THE ONYCHOPHORA

Clitellate annelids such as *Tubifex*, *Rhynchelmis*, *Glossiphonia* and *Theromyzon* illustrate the adaptation of annelid development to extreme lecithotrophy without loss of total cleavage, the formation and structure of each presumptive area of the blastula being appropriately modified as compared with those of polychaetes. Yolk onychophoran embryos, with centrolecithal cleavage and a blastoderm, can reasonably be assumed to have a presumptive area pattern as outlined above. It can now be seen that each of the presumptive areas in Onychophora has its functional equivalent in the Clitellata, and the two can be compared as follows (Figs 13, 14, 18, 58).

The *presumptive midgut* is midventral to posterior in both. In clitellates it comprises a group of large yolk cells forming the ventral wall of the blastula. There is also reason to believe that it includes a number of small posterior midgut cells cut off from the M-cells. Although these cells have not been recorded in lecithotrophic clitellate embryos, they are known to be present in albumenotrophic clitellate embryos (Tannreuther, 1915; Swetloff, 1923*b*). They also occur in certain lecithotrophic polychaete embryos in which the large, yolk, ventral midgut cells show vitellophage adaptation and the small posterior midgut cells behind them give rise to the posterior part of the midgut in the growing trunk (see Anderson, 1966*a*). Since the posterior midgut cells have the same fate in secondarily specialized albumenotrophic clitellate embryos, it seems likely that they are also present in primitive lecithotrophic clitellate embryos, in which the further development of the yolk midgut cells shows extreme vitellophage specialization (Whitman, 1878, 1887; Bürger, 1902; Vedjovsky, 1888-92; Schmidt, 1917, 1922; Penners, 1924, 1934; Iwanoff, 1928) and the development of the posterior part of the midgut in the growing trunk has not been carefully studied.

In the Onychophora, the presumptive midgut comprises a ventral sheet of blastoderm cells and a separate small group of cells posteriorly in the midline, just in front of the presumptive mesoderm.

The large, yolk cells in clitellates undergo divisions as they become internal and show vitellophage specialization, usually by segregation of small cells within a yolk syncytium, before giving rise to the major part of the midgut. Posterior midgut cells probably sink in and multiply to form the posterior part of the midgut in the growing trunk. In Onychophora, the ventral presumptive midgut cells sink slightly into the yolk and bud at the surface, producing small cells which migrate through the yolk acting as vitellophages, then accumulate at the yolk surface as the main anterior part of the midgut. The presumptive posterior midgut cells sink in slightly, then bud off a stream of cells which form the posterior part of the midgut in the growing trunk. The midgut presumptive areas in both are thus, so far as can be discerned, similar in relative position and fate, and differ only in the greater adaptation of the onychophoran areas to yolk, through (i) early segregation of cells from yolk, ventrally and posteriorly; (ii) budding of ventral cells at the surface and migration through the yolk mass; (iii) early temporary vitellophage function of the migrating cells; (iv) budding of posterior cells at the surface; (v) greater contribution by posterior cells to midgut formation, associated with greater vitellophage specialization of the ventral component in the large yolk mass anteriorly.

For the experimentalist, the critical problem of this transition is how the epigenetic system producing a large yolk cell determined as midgut and regionalized ventrally can transform into the epigenetic system producing a small yolkless cell, identically determined and regionalized, at the surface of an acellular internal yolk mass. Although several embryos in which this transition takes place during cleavage are known among arthropods, the epigenetics of such processes are still obscure.

The *presumptive mesoderm* in clitellates and Onychophora is posterior in both. In the former it comprises a pair of large M-cells which become active as teloblasts while still at the surface, budding off two lateral streams of cells, potentially metameric, which push forward on either side of the yolky developing midgut. As they bud, the teloblasts gradually sink below the surface. In Onychophora, the presumptive mesoderm comprises a group of small cells cut off at the yolk surface, which sink in slightly and bud off two lateral streams of cells, again potentially metameric, pushing anteriorly on either side of the yolk in a similar way. As with the presumptive midgut, the only essential difference is adaptation to rapid budding in spite of increased yolk, substituting segregation of budding cells from yolk reserves in place of increased teloblast size.

The *presumptive germ cells* in both clitellates and Onychophora are small cells closely associated with the presumptive mesoderm and posterior midgut, and are thus similar in location and fate.

The *presumptive stomodaeum* is also basically similar in both, comprising a group of small superficial cells at the anterior margin of the presumptive midgut. Commensurate with the slight invagination of presumptive midgut in Onychophora, the initial penetration of adjacent stomodaeum into the interior is by invagination rather than by growth and cell division, but this minor discrepancy does not belie the correspondence of the two areas.

In respect of their *presumptive proctodaeum*, clitellate and onychophoran embryos appear superficially to be different. In the former, the presumptive proctodaeum can be interpreted as a paired bilateral rudiment on either side of the presumptive mesoderm, later coming together to lie above and behind the M-cells superficially. In the Onychophora, it is a midventral rudiment between the ventral and posterior components of the presumptive midgut. Given the precocious expansion of presumptive ectoderm in Onychophora, however (see below), together with the superficial position retained by the midgut and mesodermal rudiments, the shift of presumptive proctodaeum from a paired bilateral to a median ventral position in front of the mesoderm and posterior midgut must follow. Once in this position, its initial penetration by invagination rather than by the independent ingrowth that occurs in clitellates is, like that of the stomodaeum, a corollary of its new relationship with the modified ventral presumptive midgut.

The *presumptive ectoderm* in the onychophoran blastoderm takes on directly the pattern of ectodermal bands and temporary yolk sac ectoderm found in clitellates towards the end of gastrulation. By this time, in clitellates, the ectoteloblasts have budded off ectodermal bands which run forwards ventrolaterally to meet in the anterior midline in front of the stomodaeum, retaining teloblastic activity at their posterior ends. The temporary yolk sac ectoderm has spread to cover the surface of the embryo dorsally and laterally and has also been cut off as two marginal ventrolateral strips, extending on either side from the stomodaeum to the proctodaeum between the edges of the ectodermal bands and the still-exposed surface of the midgut rudiment. In Onychophora, where the stomodaeal, midgut, proctodaeal and mesodermal areas are small and lie in line along the ventral surface of the blastoderm, a direct formation of expanded ectoderm as blastoderm takes place, omitting the gastrulation spread and early teloblastic activity of clitellate ectoderm, but arriving at an identical condition of (a) paired broad lateral ectodermal bands meeting anteriorly in front of the stomodaeum and retaining the potentiality for proliferative activity at their posterior ends. These ends lie on either side of the midventral presumptive mesoderm and presumptive posterior midgut, and just behind the now midventral presumptive proctodaeum; (b) a broad dorsal and lateral area of presumptive temporary yolk sac ectoderm; (c) paired ventrolateral bands of presumptive temporary yolk sac ectoderm, running from stomodaeum to posterior midgut between the ectodermal bands and the ventral presumptive midgut and

proctodaeum. In association with the new position of the proctodaeum, presumptive pygidium is lost.

It is manifest that in spite of great superficial dissimilarity, the basic organization of the primitive clitellate blastula and the primitive onychophoran blastoderm are similar, all the detailed differences between them being functional adaptations in Onychophora to increased yolk. Large cells containing large amounts of reserve material are replaced by small cells at the surface of a central mass of reserve material. Cells previously adapted to temporary intracellular digestion of yolk have become adapted to temporary extracellular digestion of yolk. Presumptive ectoderm, in association with a relative reduction in size of other areas, develops directly in expanded post-gastrular form. Since the Onychophora retain a basic developmental pattern so easily derivable from that of clitellate annelids, in the same way that that of clitellates is derivable from the developmental pattern of polychaetes, a "spiral cleavage" origin for the Onychophora can hardly be denied. There are also strong grounds for believing that the soft-bodied coelomates which were the ancestors of the Onychophora, Myriapods and Hexapoda (Manton, 1964, 1965) must have had a clitellate-like pattern of embryonic development. The evidence of comparative morphology, of course, argues strongly against a clitellate origin of Onychophora, and we must assume that this ancestral clitellate-like pattern of development existed in some other, now extinct, group of metameric coelomates of the spiral cleavage assemblage. By analogy with the Clitellata (see above, p. 27) it may be further assumed that the more distant ancestors of the Onychophora combined the metameric coelomate condition with a mode of development resembling that of polychaete annelids. It is important to stress, however, that no more than a paraphyletic relationship between Annelida and Onychophora can be inferred from this. Thus, while the evidence of embryology tends to link the Annelida and Onychophora in paraphyly rather than to segregate them in polyphyly, it also supports the view of Manton (1965) that "the ancestral soft-bodied coelomates from which the Arthropoda probably descended may have been quite unlike any known annelid".

References

- ANDERSON, D. T., 1959.—The embryology of the polychaete *Scoloplos armiger*. *Q. Jl. microsc. Sci.*, 100 : 89-166.
- , 1962.—The embryology of *Dacus tryoni* (Frogg.) (Diptera, Trypetidae (=Tephritidae)), the Queensland fruit-fly. *J. Embryol. exp. Morph.*, 10 : 248-292.
- , 1966a.—The comparative embryology of the Polychaeta. *Acta Zool.*, Stockh., 47 : 1-42.
- , 1966b.—The comparative embryology of the Diptera. *A. Rev. Ent.*, 11 : 23-46.
- BERGH, R. S., 1890.—Neue Beiträge zur Embryologie der Anneliden. I. Zur Entwicklung und Differenzierung des Keimstreifens von *Lumbricus*. *Z. wiss. Zool.*, 50 : 469-526.
- BOUVIER, E. L., 1904.—Les oeufs des Onychophores. *Nouv. Arch. Mus. Hist. nat.*, Paris, ser. 4, 6 : 1-50.
- BÜRGER, O., 1891.—Beiträge zur Entwicklungsgeschichte der Hirudineen. Zur Embryologie von *Nephilis*. *Zool. Jb.* (2), 4 : 697-783.
- , 1902.—Weitere Beiträge zur Entwicklungsgeschichte der Hirudeen. Zur Embryologie von *Clepsine*. *Z. wiss. Zool.*, 72 : 525-544.
- BYCHOWSKY, A., 1921.—Ueber die Entwicklung der Nephridien von *Clepsine sexoculata* Bergmann (*complanata* Savigny). *Revue suisse Zool.*, 29 : 41-131.
- DAWYDOFF, G., 1942.—Étude sur l'embryologie des Naididae indochinois. *Archs Zool. exp. gén., Notes et Revue*, 81 : 173-194.
- DENDY, A., 1902.—On the oviparous species of Onychophora. *Q. Jl. microsc. Sci.*, 45 : 363-416.
- DIMPKER, A. M., 1917.—Die Eifurchung von *Herpobdella atomaria* Carena. (*Nephilis vulgare* Moq. Tand.) *Zool. Jb.* (2), 40 : 245-290.
- EVANS, R., 1902.—On the Malayan species of Onychophora. Part II. The development of *Eoperipatus weldoni*. *Q. Jl. microsc. Sci.*, 45 : 41-86.
- HATSCHKE, B., 1878.—Studien über Entwicklungsgeschichte der Anneliden. *Arb. zool. Inst. Univ. Wien.*, 3 : 277-404.
- HOFFMANN, R. W., 1899.—Beiträge zur Entwicklungsgeschichte der Oligochäten. *Z. wiss. Zool.*, 66 : 335-57.
- IWANOFF, P. P., 1928.—Die Entwicklung der Larvalsegmente bei den Anneliden. *Z. Morph. Ökol. Tiere*, 10 : 62-161.

- KENNEL, J., 1884.—Entwicklungsgeschichte von *Peripatus Edwardsi* Blanch und *Peripatus torquatus* n. sp. *Arb. zool. zootom. Inst. Würzburg*, 7: 95–229.
- , 1885.—Entwicklungsgeschichte von *Peripatus Edwardsi* Blanch und *Peripatus torquatus* n. sp. *Arb. zool. zootom. Inst. Würzburg*, 8: 1–93.
- KOWALEVSKY, A., 1871.—Embryologische Studien an Würmern und Arthropoden. *Zap. imp. Akad. Nauk.*, 16, No. 12: 70 pp.
- MANTON, S. M., 1949.—Studies on the Onychophora. VII. The early embryonic stages of *Peripatopsis*, and some general considerations concerning the morphology and phylogeny of the Arthropoda. *Phil. Trans. R. Soc., B*, 233: 483–580.
- , 1964.—Mandibular mechanisms and the evolution of the arthropods. *Phil. Trans. Roy. Soc. B*, 247: 1–183.
- , 1965.—The evolution of arthropodan locomotory mechanisms, Part 8. Functional requirements and body design in Chilopoda, together with a comparative account of their skeleto-muscular systems and an Appendix on a comparison between burrowing forces of annelids and chilopods and its bearing upon the evolution of the arthropodan haemocoel. *J. Linn. Soc. Zool.*, 46: 251–484.
- MEYER, A., 1929.—Die Entwicklung der Nephridien und Gonoblasten bei *Tubifex rivulorum* Lam. nebst Bemerkungen zum natürlich system der Oligochäten. *Z. wiss. Zool.*, 133: 517–562.
- , 1931.—Cytologische Studien über die Gonoblasten in der Entwicklung von *Tubifex*. *Z. Morph. Ökol. Tiere*, 22: 269–286.
- NUSBAUM, J., 1884.—Zur Entwicklungsgeschichte der Hirudineen. (*Clepsine*.) *Zool. Anz.*, 7: 609–615.
- , 1885.—Zur Entwicklungsgeschichte der Geschlechtsorgane der Hirudineen (*Clepsine complanata* Sav.). *Zool. Anz.*, 8: 181–184.
- , 1886.—Recherches sur l'organogenèse des Hirudinées (*Clepsine complanata* Sav.). *Arch. Slav. Biol.*, 1: 320–340 and 539–556.
- PENNERS, A., 1922.—Die Furchung von *Tubifex rivulorum* Lam. *Zool. Jb. (2)*, 43: 323–368.
- , 1924.—Die Entwicklung des Keimstreifs und die Organbildung bei *Tubifex rivulorum* Lam. *Zool. Jb. (2)*, 45: 251–308.
- , 1929.—Entwicklungsgeschichte Untersuchungen an marinen Oligochäten. I. Furchung, Keimstreif, Vorderdarm und Urkeimzellen von *Pelosclex benedeni* Udekem. *Z. wiss. Zool.*, 134: 307–344.
- , 1934.—Die Ontogenese des entodermalen Darmepithels bei limicolon Oligochäten. *Z. wiss. Zool.*, 145: 497–507.
- , and STABLEIN, A., 1930.—Über die Urkeimzellen bei Tubificiden (*Tubifex rivulorum* Lam. und *Limnodrilus udekemianus* Claparède). *Z. wiss. Zool.*, 137: 606–626.
- SCHLIEP, W., 1914.—Die Furchung des Eies der Rüsselegel. *Zool. Jb. (2)*, 37: 313–368.
- SCHMIDT, G. A., 1917.—Zur Entwicklung des Entoderms bei *Protocleipsis tessellata*. *Ann. zool. Abt. Ges. Freunde N.A. & E.*, 4 (N.S.) Mosc., 1: 22.
- , 1921.—Die Embryonalentwicklung von *Piscicola geometra* Blainv. *Zool. Anz.*, 53: 123–127.
- , 1922.—Zur Frage über die Entwicklung des Entoderms bei der *Rhynchelmis limosella* Hoffm. *Russk. zool. Zh.*, 3: 74–93.
- , 1924.—Untersuchungen über die Embryologie der Anneliden. 2. Die Besonderheiten der Embryonalentwicklung der Ichthyobdellidae und ihre Entstehung. *Zool. Jb. (2)*, 46: 199–244.
- , 1925a.—Die polaren plasmatischen Massen Ei von *Protocleipsis tessellata*. *Russk. zool. Zh.*, 5: 138–164.
- , 1925b.—Untersuchungen über die Embryologie der Anneliden. I. Die Embryonalentwicklung von *Piscicola geometra* Blainv. *Zool. Jb. (2)*, 47: 319–428.
- , 1930.—Der Keimentwicklungstypus der *Crangonobdella murmicanica* W. D. Selenck. *Russk. zool. Zh.*, 10: 5–15.
- , 1939.—Dégénérescence phylogénétique des modes de développement des organes. *Arch. Zool. exp. gén.*, 81: 317–370.
- , 1941.—Researches on the comparative embryology of leeches. 10. Early stages of development of the embryo in Ichthyobdellidae. A la memoire A. L. Sewertzoff, Moscow, 2: 357–489.
- , 1944.—Adaptive significance of the peculiarities of the cleavage process in leeches. *Sh. obshch. Biol.*, 5: 284–303.
- SCHOUMKINE, O. B., 1953.—Embryonic development of *Hirudo*. *Trudy Inst. Morf. Zhivot.*, 8: 216–279.
- SCLATER, W. L., 1888.—On the early stages of the development of a South American species of *Peripatus*. *Q. Jl. microsc. Sci.*, 28: 343–363.
- SEDGEWICK, A., 1885.—The development of *Peripatus capensis*. Part I. *Q. Jl. microsc. Sci.*, 25: 449–468.
- , 1886.—The development of the Cape species of *Peripatus*. Part 2. *Q. Jl. microsc. Sci.*, 26: 175–212.
- , 1887.—The development of the Cape species of *Peripatus*. Part 3. *Q. Jl. microsc. Sci.*, 27: 467–550.
- , 1888.—The development of the Cape species of *Peripatus*. Part 4. *Q. Jl. microsc. Sci.*, 28: 373–398.

- SHELDON, L., 1888.—On the development of *Peripatus Novae-Zelandiae*. *Q. Jl. microsc. Sci.*, 28 : 205-237.
- , 1889a.—On the development of *Peripatus Novae-Zelandiae*. *Q. Jl. microsc. Sci.*, 29 : 283-294.
- , 1889b.—The maturation of the ovum in the Cape and New Zealand species of *Peripatus*. *Q. Jl. microsc. Sci.*, 30 : 1-29.
- SUKATSCHOFF, B., 1903.—Beiträge zur Entwicklungsgeschichte der Hirudineen. II. Über die Furchung und Bildung der embryonalen Anlagen bei *Nephilis vulgaris* Moq.-Tand. (*Herpobdella atomaria*). *Z. wiss. Zool.*, 73 : 321-367.
- SWETLOFF, P., 1923a.—Sur la segmentation de l'oeuf chez le *Bimastus constrictus* R. (fam. Lumbricidae). *Izv. biol. nauchno-issled. Inst. biol. Sta. perm. gosud. Univ.*, 1 : 101-110.
- , 1923b.—Sur la segmentation de l'oeuf de *Rhynchelmis limosella* Hoffnstr. *Izv. biol. nauchno-issled. Inst. biol. Sta. perm. gosud. Univ.*, 1 : 141-152.
- , 1926.—Über die Embryonalentwicklung bei den Naididen. *Izv. biol. nauchno-issled. Inst. biol. Sta. perm. gosud. Univ.*, 4 : 359-372.
- , 1928.—Untersuchungen über die Entwicklungsgeschichte der Regenwürmer. *Trudy osob. zool. Lab. sebastop. biol. Sta.*, 13 : 95-329.
- TANNREUTHER, G. W., 1915.—The early embryology of *Bdellodrilus philadelphicus*. *J. Morph.*, 26 : 143-216.
- VEDJOVSKY, F., 1886.—Die Embryonalentwicklung von *Rhynchelmis*. *Sber. K. boh. Ges. Wiss.*, 2 : 227-239.
- , 1888-1892.—Entwicklungsgeschichtliche Untersuchungen. Prag.
- WHITMAN, C. O., 1878.—The embryology of *Clepsine*. *Q. Jl. microsc. Sci.*, 18 : 215-315.
- , 1887.—A contribution to the history of the germ layers of *Clepsine*. *J. Morph.*, 1 : 105-182.
- WILSON, E. B., 1889.—The embryology of the earthworm. *J. Morph.*, 3 : 387-462.
- , 1892.—The cell lineage of *Nereis*. *J. Morph.*, 6 : 361-480.